

Chapter 6

Concentrations and Human Health Risks Posed by Chemicals in the Tissue of Black Mussels and West Coast Rock Lobsters

6.1. Introduction

A critical end-point of any outfall monitoring programme is to determine if effluent discharge is significantly adversely impacting on the ecology of the receiving environment and/or is posing a risk to the health of humans that use that environment. It is not possible to determine if chemical pollution has occurred, and the extent and significance of the pollution, either alone or relative to other stressors inducing ecosystem alteration, by simply measuring concentrations of chemicals in the environment (Ludwig and Iannuzzi, 2005). These measurements provide information on contamination, that is, chemicals present where they would not normally occur or at concentrations above the natural background, but do not provide information on pollution, that is, contamination that causes adverse biological effects in the environment. All pollutants are contaminants, but not all contaminants are pollutants since chemicals introduced into the environment may be more or less bioavailable to organisms depending on their form, modifying factors in the environment, the environmental compartment they occupy, and the reactions (behavioural and physiological) of exposed biota (Chapman *et al.*, 2003). Determining if contamination has resulted in pollution thus requires chemical and biological measurements.

There are numerous biological indicators that can be used to evaluate the ecological impact of effluent discharge. As discussed in Chapter 5 the most widely used approach in marine effluent discharge monitoring programmes is to compare the structure and composition of benthic macrofaunal communities between putatively effluent impacted and reference sites. Aberrations in the type and abundance of organisms at sites near the point of effluent discharge relative to

communities at distant sites are taken as being indicative of an impact provided there are no factors confounding such interpretation. There are also certain benthic macrofaunal organisms that thrive in sediment significantly impacted by effluent discharge (*e.g.* capitellid polychaetes) and these are widely used as indicators of impact. The challenge of using benthic macrofauna for this purpose in the Cape Town outfalls monitoring programme was discussed in Chapter 5. To reiterate, the presence of exposed reef near the Green Point and Camps Bay outfalls made it difficult to impossible to collect sediment for benthic macrofaunal community analysis in areas putatively most impacted by effluent discharge through these outfalls.

A possible fate for contaminants in the marine environment is their bioaccumulation by marine fauna and flora. The contaminant burden may affect the health of marine organisms and the health of humans that consume fauna and flora from the marine environment. The significance is that even if concentrations of chemicals in effluent are compliant with license conditions and/or are diluted to below water quality targets for ecosystem protection near the point of discharge, certain chemicals that may be in effluent may still pose an ecological and human health risk through the processes of bioaccumulation and biomagnification. Bioaccumulation is the process of uptake and retention of chemicals in marine fauna and flora. Biomagnification describes the progressive increase in a chemical's concentration through successive trophic levels of the food web, with the highest concentrations found in apex predators (*e.g.* in sharks and large predatory fish such as tuna).

For the 2015/2016 survey of the Cape Town outfalls

monitoring programme, mussels and rock lobsters were identified as indicators of effluent discharge impacts for two reasons. First, mussels and rock lobsters in other parts of South Africa and/or the world are known to accumulate contaminants in their tissue. Second, mussels and rock lobsters are targeted by commercial and recreational fishers in the Cape Town outfalls study area. The objective was, therefore, to use mussels and rock lobsters as contaminant monitoring sentinels for, and indicators of potential health risks posed to humans through seafood consumption due to effluent discharge through the Green Point, Camps Bay and Hout Bay outfalls.

So-called Mussel Watch programmes¹, wherein bivalves are used as contaminant monitoring sentinels have been implemented in many parts of the world, including South Africa (e.g. Monirith *et al.*, 2002; Andral *et al.*, 2004; Liu and Kueh, 2005; Mubiana *et al.*, 2005; Wei *et al.*, 2006; Isobe *et al.*, 2007; Kimbrough *et al.*, 2008; CSIR, 2016c). The basis for using bivalves is their ability to bioaccumulate contaminants in their tissue to a degree that is proportional to the contaminants bioavailability. Many contaminants that bivalves accumulate also undergo minimal metabolic transformation in their tissue, unlike in other organisms such as fish, where substantial metabolic transformation may occur. This and the fact that bivalves are sessile make them good indicators of 'recent' (one to a few months) local contamination of surrounding waters (Roesijadi *et al.*, 1984; Sericano *et al.*, 1995; Kimbrough *et al.*, 2008). There are other reasons bivalves are useful contaminant monitoring sentinels. First, they are usually widely distributed, albeit not necessarily the same species. This makes them valuable indicators of local conditions and for broader-scale comparison when collected from multiple locations. Second, they are usually abundant and easy to collect and identify. Third, they bioaccumulate contaminants in their tissue to far higher concentrations than in the water column. This makes the measurement of contaminant concentrations in the laboratory

easier. Fourth, contaminant concentrations in the water column are usually highly variable due to turbulence/mixing (e.g. currents) and stochastic anthropogenic inputs. The analysis of water samples thus provides a snapshot of coastal water quality at the time of monitoring and important contamination events may be missed. Bivalves, in contrast, provide a more temporally integrated measure of contamination since they achieve equilibrium with the surrounding water and take time to depurate contaminants.

Rock (and other) lobsters are less commonly used as contaminant monitoring sentinels. However, because many contaminants adhere to particulate organic matter in effluent and many marine organisms that live on the seabed will consume this matter when it settles from the water column, these organisms (through their prey), and in turn organisms that consume them are susceptible to the toxic effects of contaminants bioaccumulated in their tissue.

This chapter discusses the findings of the chemical analysis of the tissue of mussels and rock lobsters collected at various sites along the Cape Peninsula for the 2015/2016 survey of the Cape Town outfalls monitoring programme and the potential risk posed by chemicals in their tissue to human consumers.

6.2. Materials and Methods

6.2.1. Fieldwork and Laboratory Analyses

About 40 black mussels (*Mytilus galloprovincialis*, often referred to as the Mediterranean mussel) were collected by hand from the rocky intertidal at eight sites between Mouille Point and Kommetjie on the Cape Peninsula in 2016 (Figure 6.1). West coast rock lobsters (*Jasus lalandii*) were collected using baited traps deployed in the nearshore marine environment at seven sites between Green Point and Hout Bay (Figure 6.2). Rock lobster tails were also purchased in a store. The mussels and rock lobsters were wrapped in aluminium foil and placed on ice in the field, and frozen on return to the laboratory. Mussels and rock lobsters were collected as per conditions stipulated in permit RES2016/92 issued to the City of Cape Town jointly by the Department of Environmental Affairs and Department of Agriculture, Fisheries and Forestry.

¹ While the term Mussel Watch is used to describe this type of monitoring, mussels are not the only organisms used as sentinels. Oysters are widely used for this purpose, because mussels are not always present at all locations of interest and/or because mussels and oysters accumulate chemicals/contaminants to varying degrees in their tissue.

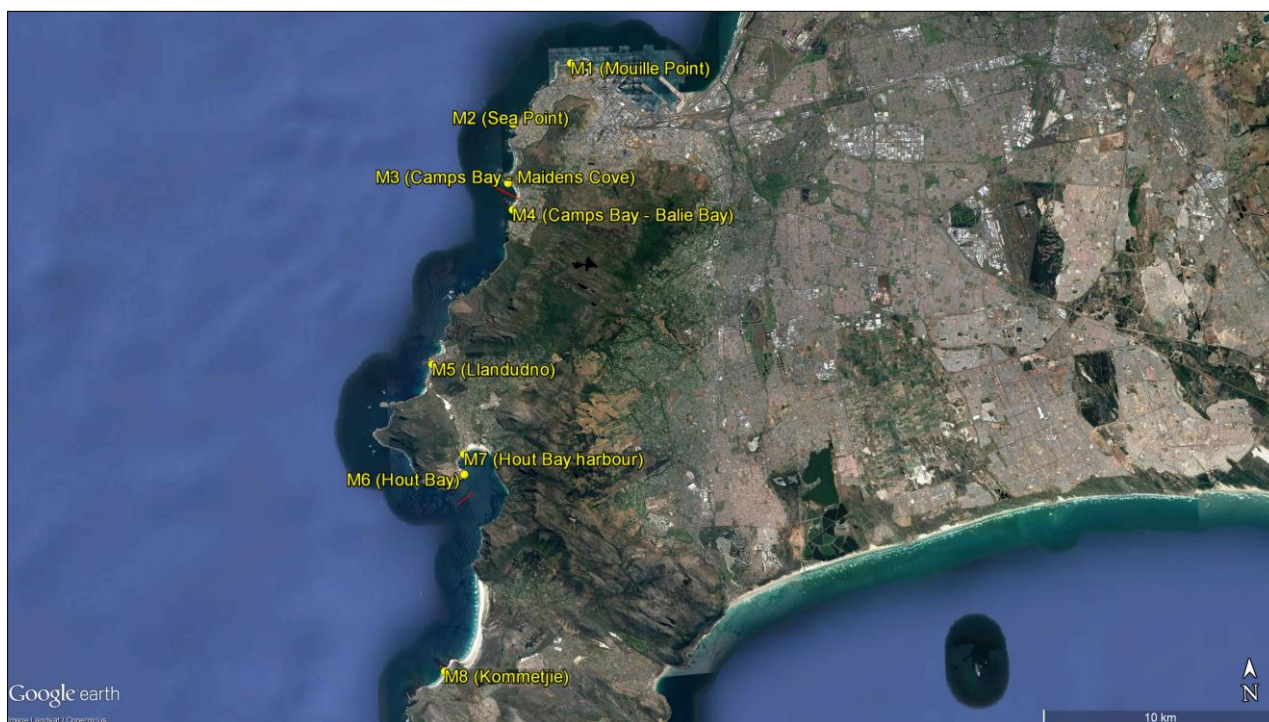


Figure 6.1. Aerial view of the Cape Peninsula area of Cape Town, showing the positions where black mussels (*Mytilus galloprovincialis*) were collected in 2016. The outfalls are denoted by red lines.

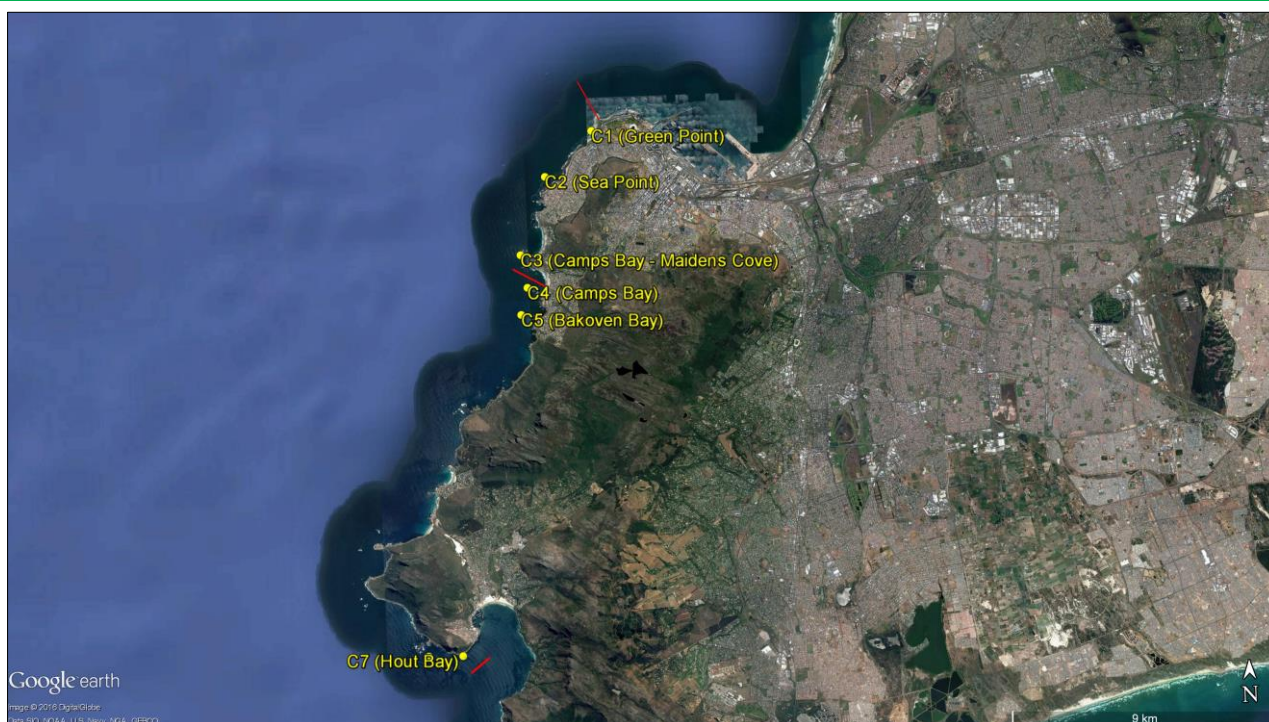


Figure 6.2. Aerial view of the Cape Peninsula area of Cape Town, showing the positions where west coast rock lobsters (*Jasus lalandii*) were collected in 2016. The outfalls are denoted by red lines.

M. galloprovincialis is a non-native mussel that was introduced to South Africa in the late 1970's (Branch and Steffani, 2004). Natural spread from the west coast has resulted it now having a range from central Namibia to just beyond East London (Robinson *et al.*, 2005; Mead *et al.*, 2011). *M. galloprovincialis* is a filter feeder, meaning it pumps water across its gills that have specialised

structures that remove small particles of suspended organic matter and plankton from the seawater. Stable isotope analysis of *M. galloprovincialis* tissue by Bustamante and Branch (1996) revealed that on the west coast of South Africa they obtain 60 - 70% of their nitrogen and carbon from kelp-derived particles. Phytoplankton constitutes most of the remaining 30 - 40%.

The panulirid west coast rock lobster *J. lalandii* is a commercially exploited lobster that has a range from Namibia to about East London. It is typically associated with rocky reefs and its predation strongly influences the abundance and population structure of its prey (Barkai *et al.*, 1996), which includes coralline algae, mussels, urchins, starfish, barnacles and fish (Mayfield *et al.*, 2000).

6.2.2. Mussel and Rock Lobster Processing

In the laboratory, mussels were partially thawed and their total length (anterior umbo to posterior growing lip) measured using Vernier callipers. Mussels between 75 - 85 mm total length were targeted, although ten mussels across the sites of 70.2 - 74.2 mm were analysed. The mussels were then shucked in a partially thawed state, their byssus threads trimmed, and their individual wet tissue weight measured to the nearest 0.1 g. The tissue of between 11 - 15 mussels (depending on their weight) was composited in pre-weighed acid and solvent rinsed glass jars with foil lined lids. The weight of the sample container after tissue addition was determined. The tissue was then freeze dried and the container weight measured. The difference in container weight before and after freeze drying was used to determine tissue moisture content. The dried tissue was ball milled and then passed through a 1 mm mesh size sieve to remove material that could not be milled to a fine consistency (*e.g.* adductor muscle).

Rock lobsters were partially thawed, their total, carapace and tail length measured using Vernier calipers, and their wet weight determined to the nearest 0.1 g. The tail was then dissected, the intestinal tract removed, and the abdominal muscle tissue removed and cut into small cubes. The tissue of three (but in one case two) rock lobsters with a broadly comparable tail length per site was composited in pre-weighed acid and solvent rinsed glass jars with foil lined lids. Tissue processing (*i.e.* wet weight determination, freeze drying, sieving) was done in the same manner for described above for mussels.

Five rock lobster tails purchased from a store (source unknown) were processed in the same manner as field collected specimens, but here only the tail length and wet weight were measured.

All apparatus used to clean, shuck, dissect and macerate the tissue of mussels and rock lobsters was comprised of non-contaminating material (*e.g.* stainless steel knives), and was cleaned between the processing of mussels and rock lobsters from different sites by sequentially rinsing in 10% nitric acid, hexane and deionised water.

6.2.3. Laboratory Analyses

6.2.3.1. Chemicals Identified for Analysis²

Concentrations of metals, polycyclic aromatic hydrocarbon isomers, polychlorinated biphenyl congeners and non-steroidal anti-inflammatory drugs were analysed in the tissue of mussels and rock lobsters. These chemicals were targeted for several reasons.

First, various metals and polycyclic aromatic hydrocarbon isomers were consistently detected in effluent collected from the Green Point, Camps Bay and Hout Bay wastewater works in 2016, often at concentrations higher than can be expected to occur naturally in seawater in the marine receiving environments. As discussed in Chapter 5, several metals and polycyclic aromatic hydrocarbons were also common contaminants of sediment in the Table Bay and to a lesser degree Camps Bay and Hout Bay areas (Hout Bay Harbour excluded).

Second, non-steroidal anti-inflammatory drugs are more or less ubiquitous constituents of effluent and thus present a more specific chemical marker of effluent than metals and polycyclic aromatic hydrocarbons, which occur naturally in the marine environment. Although metals and polycyclic aromatic hydrocarbons are not specific markers of effluent they were targeted as there was no certainty non-steroidal anti-inflammatory drugs would be detected in mussels and rock lobsters.

Third, metals, polycyclic aromatic hydrocarbons and polychlorinated biphenyls are known to be accumulated by black mussels in South Africa (*e.g.* Sparks *et al.*, 2014) and other parts of the world. If these chemicals are at elevated concentrations in mussel and rock lobster food/prey in the greater vicinity of the Green Point, Camps Bay and Hout Bay outfalls as a consequence of effluent discharge

² Chemical concentrations are presented on a wet weight basis unless otherwise stated.

then they should theoretically be accumulated by mussels and rock lobsters.

As discussed in Chapter 5, there is a strong possibility that metals and polycyclic aromatic hydrocarbons are also introduced into the marine environment off Cape Town via stormwater and river runoff. It was for this reason polychlorinated biphenyls were analysed. As discussed in Chapter 2, poly-chlorinated biphenyls were never detected in effluent collected from the Green Point, Camps Bay and Hout Bay wastewater works in 2016, but are known contaminants of sediment in wetlands, rivers and estuaries in the greater Cape Town area (CSIR, 2015) and in sediment in the Port of Cape Town (CSIR, unpublished data). They were also detected in three sediment samples collected in Table Bay for this survey. Although the source/s of the polychlorinated biphenyls in sediment in the wetlands, rivers and estuaries and in the Port of Cape Town is unknown there is a strong probability they are entering these systems via stormwater. If so, their presence in the tissue of mussels and rock lobsters may provide a marker for contaminant uptake via exposure to stormwater and river runoff.

Lastly, certain metals, polycyclic aromatic hydrocarbons and polychlorinated biphenyls in mussels and rock lobsters are known or strongly suspected carcinogens, and/or are known to pose other health risks to humans. The consumption of mussels and rock lobsters collected on the Cape Peninsula that have accumulated these chemicals in their tissue as a result of exposure to effluent or other land-based sources of contaminants may thus pose a risk to human health.

6.2.3.2. Metal Analysis

Approximately 1 g of dried mussel or rock lobster tissue was transferred to high-pressure microwave extraction vessels containing 10 ml of HNO_3 . The tissue was digested with microwave assistance. The digestate was filtered, diluted to volume, and the concentrations of metals were detected and quantified using Inductively Coupled Plasma Mass Spectrometry and Inductively Coupled Plasma-Atomic Emission Spectrometry, or using a Direct Mercury Analyser for mercury. Standard reference material (DORM 2 - dogfish muscle; NRCC Canada)

was analysed with batches of mussel and rock lobster tissue for quality assurance purposes.

6.2.3.3. Organic Chemical Analysis

Polycyclic aromatic hydrocarbons: About 0.75 g of freeze dried and ball milled mussel or lobster tissue were mixed with 10 g of diatomaceous earth and relevant isotope labelled internal standards were added. The samples were extracted by means of an accelerated solvent extractor (three cycles) using a hexane:acetone mixture (1:1 v/v) at 100°C and 1 500 psi. The extracts were dried with anhydrous sodium sulphate and evaporated until almost dry using a rotary evaporator. The residues were reconstituted in 15 ml ethyl acetate:cyclohexane (1:1 v/v) and 5/6 of the extracts were purified by means of gel permeation chromatography. The fractions were evaporated till dryness using silanized tubes and then reconstituted in hexane and purified over an alumina column. The extracts were again evaporated till dryness using silanized tubes and reconstituted in an isooctane solution. The extracts were analysed by means of gas chromatograph high resolution mass spectrometry.

Polychlorinated biphenyls: About 1.5 g of freeze dried and ball milled mussel or lobster tissue was mixed with 15 g of anhydrous sodium sulphate. All material was transferred to a 50 ml polypropylene tube and an internal standard added. After adding 25 ml of ethyl acetate the mixture was extracted for 30 minutes using a head-over-head mixer. After centrifuging for 10 minutes at 3000 rpm, 10 ml of the upper layer was reduced to 1.0 ml at 40°C under nitrogen. Next, 0.5 ml extract was purified by means of gel permeation chromatography and the collected fractions were reduced to 0.5 ml at 40°C under nitrogen. The resulting cleaned extract was analysed by means of gas chromatograph triple quad mass spectrometry.

Non-steroidal anti-inflammatory drugs: About 2 g of freeze dried and ball milled mussel or lobster tissue was transferred into a 50 ml polypropylene tube. Five ml of acetonitrile was added and the extract was mixed for 15 minutes using a head-over-head mixer. The tubes were centrifuged for 10 minutes at 3500 rpm. The supernatant was transferred to a 12 ml polypropylene tube containing 200 mg of primary-secondary amine.

The tubes were mixed for 5 minutes using a head-over-head mixer followed by centrifuging for 10 minutes at 3500 rpm. The supernatant was transferred into a 12 ml glass tube and evaporated at 55°C under nitrogen. The dried extract was reconstituted in 800 µl methanol and analysed by means of liquid chromatography triple quad mass spectrometry.

6.4. Risk assessment

6.4.1. General

Risk assessment is a process by which the degree and nature of a risk is characterised. The outcome determines if there is a need for risk management, that is, prevention and control measures to reduce the risk. In the context of seafood this could include an advisory on the consumption of fish and shellfish caught or collected in a particular area due to high contaminant concentrations in their tissue in addition to measures for the identification, control and reduction of contaminants at their source.

The risk assessment approach followed in this study is identical to that recommended by the United States Environmental Protection Agency (USEPA, 2000 b) for evaluating risks arising to human health by exposure to chemicals in fish and shellfish. The risk assessment comprises four stages, namely:

- Hazard identification,
- Dose-response assessment,
- Exposure assessment,
- Risk characterisation.

6.4.2. Hazard Identification

Hazard identification determines if exposure to a chemical can increase the incidence of a particular adverse health effect and the likelihood of such an occurrence. Screening Values, which represent concentrations of chemicals in fish and shellfish of potential human health concern, were calculated for two components of the South African population, namely low- and high-end consumers (these terms are discussed further below). Screening Values for non-carcinogenic and carcinogenic risks were calculated, at a risk level of 1×10^{-5} for the latter. Concentrations of inorganic arsenic in rock lobsters collected at most sites and polycyclic aromatic hydrocarbons in mussels collected at one site exceeded Screening Values for carcinogenic risk

(0.0037 mg.kg⁻¹ for inorganic arsenic and 0.0008 mg.kg⁻¹ for polycyclic aromatic hydrocarbons) as per the high-end consumer scenario, while the cadmium concentration in mussels collected at one site exceeded the Screening Value (0.558 mg.kg⁻¹) for non-carcinogenic risk as per the high-end consumer scenario. This identified the need for a detailed assessment of potential health risks as a result of exposure to these chemicals through a seafood consumption pathway.

6.4.3. Dose-Response Assessment

The quantitative relationship between a chemical dose and the incidence of carcinogenic and non-carcinogenic health effects in humans was assessed using toxicity data from the United States Environmental Protection Agency Integrated Risk Information System.

6.4.4. Exposure Assessment

The goal of exposure assessment is to identify populations that might be exposed to chemicals, the pathway through which they might be exposed, and the variables for exposure assessment that allow the chemical dose to be quantified.

6.4.4.1. Identification of Exposed Populations

As mentioned above two exposed populations were identified, namely low- and high-end consumers. These are distinguished by the amount of seafood they consume, based on information on fish consumption by South Africans provided by Steyn *et al.* (2003) (see further below).

The United States Environmental Protection Agency (USEPA, 2000a,b) recommends the consideration of sensitive and insensitive segments of exposed populations. Sensitive segments are defined as infants, children and females of childbearing age, while insensitive segments are adult males and adult females beyond their childbearing years. These were not considered separately for this risk assessment since consumption rate was taken as proportional to body weight. Assessing the risk to an adult thus concurrently caters for the risk to females of childbearing age, children and infants (but with some caveats).

6.4.4.2. Exposure Pathway

Although several pathways could conceivably result in human exposure to a chemical of concern, for this risk assessment the consumption of mussels and rock lobsters was considered the only pathway of exposure. This is an obvious simplification of the real-world situation since many foodstuffs contain metals and polycyclic aromatic hydrocarbons, amongst other chemicals. Other pathways besides consumption, such as inhalation, may also result in exposure.

6.4.4.3. Quantification of Exposure

An individual's exposure to contaminants through a seafood consumption pathway depends on several factors, such as the concentration of contaminants in seafood, the amount of seafood consumed, how often and for how long seafood is consumed, and the consumer's body weight. Because exposure occurs over time the total exposure is divided by a time period of interest to obtain an average exposure rate per unit time. When this is expressed as a function of body weight it is referred to as the Chemical Specific Daily Intake (CDI). The Chemical Specific Daily Intake of chemicals in mussels and rock lobsters analysed for this study was calculated as:

$$CDI = (C \times CR \times EF \times ED) / (BW \times AT) \quad \text{Equation 1}$$

Where:

CDI = Chemical Specific Daily Intake (mg.kg⁻¹.day),

C = Chemical concentration in mussel or rock lobster tissue (mg.kg⁻¹ wet weight),

CR = Consumption rate (kg.day⁻¹),

EF = Exposure frequency (days),

ED = Exposure duration (years),

BW = Body weight (kg),

AT = Averaging time (days).

6.4.4.4. Chemical Concentrations

The concentrations of chemicals analysed in mussel and rock lobster tissue (variable C in Equation 1) are provided in appendices to this report. Laboratory methods and instruments do not allow accurate measurement of chemicals below a certain concentration that is specific to the method and instrumentation used. This is known as the method detection limit. Because there is no certainty a chemical reported as below the method

detection limit was not present in the tissue of mussels and rock lobsters a decision must be made on how to treat non-detects. The most common approach is to replace non-detects with a surrogate concentration equal to one-half the method detection limit. For the purposes of this study a more conservative approach was followed, by replacing non-detects with a concentration equal to the method detection limit. This is conservative since it will provide a slightly higher estimate of risk than a surrogate concentration of one-half the method detection limit.

Arsenic is present in the tissue of marine fish and shellfish in both inorganic and organic forms, but only the inorganic form is toxic to humans (USEPA, 2000). Analysing for inorganic arsenic is complicated and expensive. Marine Chemistry Laboratories at the CSIR thus follow the same approach as many laboratories by analysing for total arsenic and assuming a scientific literature defined proportion is in an inorganic form. The literature suggests that inorganic arsenic usually comprises <2% (and often <1%) of the total arsenic concentration in marine shellfish (e.g. Greene and Crecelius, 2006; Schoof and Yager, 2007; Goessler *et al.*, 1997; Donohue and Abernathy, 1999; Schoof *et al.*, 1999a,b; Morrissey and Abernathy, 1999; De Gieter *et al.*, 2002; Li *et al.*, 2003; Fabris *et al.*, 2006; Peshut *et al.*, 2008). A 2% adjustment factor was used to estimate the proportion of inorganic arsenic in the tissue of mussels and rock lobsters collected for this study.

Mercury is present in marine fish and shellfish tissue in two predominant forms, namely elemental mercury and methylmercury. The most toxic form is methylmercury. Analysing for methylmercury is complicated and expensive and Marine Chemistry Laboratories at the CSIR follow the same approach as many laboratories by analysing for elemental mercury and assuming all of this is present as methylmercury. This approach is valid in that the contribution of methylmercury to the total mercury concentration in marine fish and shellfish is usually >90% (>95% - Bloom, 1992; 90 to 100% - USEPA 2000a, 2009; 98% - Hammerschmidt and Fitzgerald, 2006; >95% - Senn *et al.*, 2010), although the contribution can be variable (45 to 124% - Kannan *et al.*, 1988; 43 to 76% - Forsyth *et al.*, 2004; 60 to

100% - Storelli *et al.*, 2005). Assuming that all mercury is present as methylmercury is thus a conservative approach.

A number of polycyclic aromatic hydrocarbon isomers were detected in mussel and rock lobster tissue. Of these only benzo(a)pyrene has a Cancer Slope Factor (this term is discussed below). To estimate the risk of exposure to polycyclic aromatic hydrocarbons, Relative Potency Estimates derived by the USEPA (1993) based on mouse skin carcinogenesis were used to derive a summed potency for these chemicals.

The USEPA (2002) provides an Oral Reference Dose and Cancer Slope Factor for polychlorinated biphenyls that can be measured as Aroclors or congeners. Considering there are 209 possible congeners, but only seven were analysed in the tissue of mussels and rock lobsters for this study the total polychlorinated biphenyl concentration may provide an underestimate of the risk posed by these chemicals.

6.4.4.5. Consumption Rate

The consumption rate (variable CR in Equation 1) is critical for calculating the Daily Intake. Steyn *et al.* (2003) provide average per capita fish (fresh and canned) consumption rates for South Africans >10 years of 11.93 or 15.35 g per day depending on the approach used. However, only a small proportion of study participants reported they consume fish. If these persons only are considered the consumption rates are 113.46 or 125.44 g per day. For this risk assessment the consumption rate for low-end consumers was taken as the higher of the average per capita consumption rates (15.35 g per day) while the consumption rate for high-end consumers was taken as the higher of fish consumer rates (125.44 g per day).

Since consumers find it difficult to determine the rate at which they consume food it is worthwhile placing the consumption rates into context. The typical weight of (frozen) packaged fish fillets (*e.g.* hake) in stores in South Africa is about 100 - 120 g (as per package specifications). A standard can of tuna weighs 170 g including liquid (oil or water; as per package specifications) and about 120 g after draining the liquid. In South African restaurants a

fish serving is usually about 180 - 280 g in wet weight (as per restaurant menus).

Steyn *et al.* (2003) recommend using an average meal portion size for evaluating exposure of the South African population to contaminants that may be found in foods. For fish this is 140 g (note the 97.5th percentile meal size is 400 g). At a meal size of 140 g the consumption rate equates to a little more than three meals a month for low-end consumers and about 27 meals per month for high-end consumers.

6.4.4.6. Chemical Absorption

It was assumed the entire concentration of chemicals in mussel and rock lobster tissue is absorbed across the intestinal tract.

6.4.4.7. Exposure Frequency

An exposure frequency of 365 days per year was assumed for the chemical specific Daily Intake calculation, a standard practice for human health risk assessment.

6.4.4.8. Exposure Duration

The exposure duration is the period over which an individual is exposed to contaminants in seafood at the specified concentration and ingestion rate. The period low- and high-end consumers may consume mussels and rock lobsters collected in the Cape Town area is unknown. As far as the scientists that prepared this report are aware there are no population mobility studies for South Africa. The typical exposure period assumed for health risk assessment is 30 years, which is near the upper length of time (90th percentile) that residents (in fact farmers) in the United States of America spend in a residence (USEPA, 2011) and are assumed to catch and consume fish and shellfish from the same area. This exposure period was used for low- and high-end consumers.

6.4.4.9. Averaging Time

As discussed earlier, exposure to contaminants in seafood occurs over time. Therefore, the total exposure is divided by the time period of interest to obtain an average exposure rate per unit time. When this rate is expressed as a function of body weight the resulting exposure rate is referred to as

the Daily Intake expressed in milligrams of a chemical taken into the body per kilogram body weight per day.

Bradshaw *et al.* (2012) estimated the average life expectancy at birth for males and females in South Africa in 2011 at 57.2 and 62.8 years respectively. This provides an average life expectancy for South Africans of 60 years. Since a large proportion of South Africans can be expected to have a lifespan exceeding 60 years a life expectancy of 70 years was used in this risk assessment. Further motivation for using this exposure period is that the United States Environmental Protection Agency's Integrated Risk Information System (IRIS) assumes a 70 year lifetime for the derivation of cancer slope factors. The use of a 70 year life expectancy thus avoids the need to adjust cancer slope factors to a shorter life expectancy. The averaging time for estimating carcinogenic risk was thus 25 550 days, the number of days in a 70 year exposure period.

6.4.4.10. Body Weight

The South African Demographic and Health Survey for 2003 (DOH, 2007) provides the average bodyweight for South African's of 15 years and older at 66 kg for males and 68 kg for females. The average body weight is thus 67 kg. The South African National Health and Nutrition Examination Survey (Shisana *et al.*, 2013) provides an average bodyweight for South African's 15 years and older of 67.3 kg for males and 72.2 kg for females. The average body weight is thus 69.8 kg. For the purposes of this risk assessment an average body weight of 70 kg was used as it provides a conservative estimate of risk. The United States Environmental Protection Agency's Integrated Risk Information System (IRIS) also assumes a 70 kg adult body weight for the derivation of Cancer Slope Factors (this default weight for human health criteria has been revised to 80 kg for adults 21 years and older, but with no associated change in the cancer slope factors; USEPA, 2011).

6.4.4.11. Cooking Loss of Contaminants

Cooking can result in the loss of some chemicals from fish and shellfish tissue (*e.g.* Armbruster *et al.*, 1987; Zabik *et al.*, 1996; Salama *et al.*, 1998), with a concomitant reduction in risk. This is significant for organic chemicals, such as polychlorinated

biphenyls, which are usually associated with lipids commonly lost during cooking. Chemicals may also be volatilised during cooking. However, cooking loss will not result if a stew-type meal is prepared, that is, the lost lipids are not allowed to 'escape'. The situation is slightly different for metals, which can become concentrated in fish and shellfish tissue due to fluid loss (*e.g.* Ersoy *et al.*, 2006), although their bioaccessibility may decrease. This is in spite of the fact that a significant proportion of the metal content in fish and shellfish may be lost during the cooking process, although this is metal specific (Metian *et al.*, 2009). However, several workers have reported no or little loss of contaminants and even a slight increase for eels and fish during cooking (*e.g.* Trotter *et al.*, 1989; Moya *et al.*, 1998; Visciano *et al.*, 2013).

Cooking loss was not incorporated into this risk assessment given the incomplete information on how each chemical is affected by cooking. This is the most conservative approach and is in line with United States Environmental Protection Agency (USEPA, 2000a,b) guidance on fish and shellfish consumption advisories, which recommends that cooking loss should be considered only if there is information on how methods of preparation influence chemical concentrations in seafood. However, some agencies recommend reducing chemical concentrations by up to 50% for polychlorinated biphenyls and similar chemicals that have a high octanol/water partition coefficient ($K_{ow} > 3$) and are thus concentrated in fatty rather than muscle tissue.

6.4.5. Risk Characterisation

Risk characterisation integrates the results of the exposure assessment with chemical toxicity information to derive estimates of risk. Non-carcinogenic and carcinogenic risk estimates are calculated separately because of fundamental differences in their critical toxicity values.

6.4.5.1. Non-Carcinogenic Risk

In general humans that consume contaminated seafood are exposed to low concentrations of chemicals over an extended period. This type of exposure rarely results in acute toxicity, that is, exposure to a single high dose of a chemical. However, long-term exposure may result in chronic

toxicity. The potential for chronic, non-carcinogenic health effects was thus evaluated by calculating the ratio of chemical exposure to an Oral Reference Dose (RfD). This ratio of exposure to toxicity for an individual chemical, referred to as a Hazard Quotient (HQ), was calculated as:

$$HQ = CDI/RfD \quad \text{Equation 2}$$

Where:

HQ = Chemical specific hazard quotient (unitless),

CDI = Chemical specific daily intake ($\text{mg.kg}^{-1}\text{-day}$),

RfD = Chemical specific reference dose ($\text{mg.kg}^{-1}\text{-day}$).

The oral reference dose is an estimate, with an uncertainty spanning perhaps an order of magnitude (a tenfold difference), of the daily oral exposure of a population, including sensitive subpopulations, to a potentially hazardous material that is likely to be without an appreciable risk of deleterious non-carcinogenic effects over a lifetime (USEPA, 2000b). The underlying assumption of a reference dose is that there is a threshold dose below which there is a negligible risk that certain toxic effects will occur.

Because of uncertainty associated with toxicity data, 'safety factors' are included, resulting in a lower and more protective reference dose. If a Hazard Quotient exceeds a value of one (*i.e.* exceeds the Oral Reference Dose) then individuals may be at risk. The magnitude of the risk can be inferred from the degree to which the reference dose is exceeded. If the Hazard Quotient is only slightly above a value of one then the dose will likely fall below the toxic effect level because of the abovementioned safety factors. However, a Hazard Quotient is not linear, with the result that a Hazard Quotient of four does not imply a four times greater risk compared to a Hazard Quotient of one. Rather, the United States Environmental Protection Agency (USEPA, 2000b) suggests that a Hazard Quotient of less than one should be interpreted as 'no cause for concern' whereas a Hazard Quotient exceeding one should indicate some cause for concern. According to the USEPA (1989), for Hazard Quotients from 1 - 10 there is some risk of an adverse health effect occurring but this is typically within the bounds of uncertainty while a Hazard

Quotient >10 indicates a moderate to high risk of adverse health effects depending on the magnitude of the quotient.

Some investigators estimate the cumulative potential for non-carcinogenic effects due to simultaneous exposure to multiple chemicals in fish and shellfish using a Hazard Index, which is the sum of Hazard Quotients for all chemicals. However, the assessment of additive risk is only relevant if the risk is the same for all chemicals. This is not the case for chemicals analysed in mussels and rock lobsters for this study. For example, exposure to polychlorinated biphenyls may cause neuro-behavioural changes while exposure to mercury and arsenic may cause hand tremors and/or memory loss, and hyper-pigmentation respectively (USEPA, 2010). As these effects are different, non-carcinogenic risk was assessed only on the basis of the Hazard Quotient.

6.4.5.2. Carcinogenic Risk

The potential health risk posed by chemicals identified as (probable) carcinogens was estimated as the incremental probability that an individual will develop cancer over his or her lifetime as a result of chemical exposure. The term 'incremental' implies this risk corresponds to the added probability of cancer above the background cancer risk experienced by all individuals in the course of daily life. Cancer risks are expressed as a unitless probability (*e.g.* 1 in 1 million, or 1×10^{-6}) of an individual developing cancer over a lifetime above the background risk as a result of the exposure. The United States Environmental Protection Agency (USEPA, 2000b) assumes a threshold dose does not exist for carcinogens and any dose can contribute to carcinogenic health risk. In other words, there is never a zero probability of cancer risk when exposed to carcinogenic chemicals. Carcinogenic risk was calculated as an Excess Cancer Risk (ECR), as:

$$ECR = CDI \times CSF \quad \text{Equation 3}$$

Where:

ECR = Excess Cancer Risk (unitless),

CDI = Chemical specific daily intake ($\text{mg.kg}^{-1}\text{-day}$),

CSF = Chemical specific cancer slope factor ($\text{mg.kg}^{-1}\text{-day}$)

The Cancer Slope Factor is an upper-bound estimate, approximating 95% confidence limits, of the probability an individual will develop cancer over a lifetime as a consequence of exposure to a given dose of a specific carcinogen (USEPA, 2000b). Current regulatory practice suggests there is no 'safe dose' of a carcinogen and that a very small dose of a carcinogen will give a very small cancer risk. Cancer risk estimates are, therefore, not yes/no answers, but measures of probability. Such measures, however uncertain, are useful in determining the magnitude of a cancer risk because any level of a carcinogenic contaminant carries an associated risk. The interpretation of Excess Cancer Risk thus requires that an acceptable increase in cancer risk be defined. This is referred to as the acceptable risk level. There is no universally accepted acceptable risk level. The United States Environmental Protection Agency (USEPA, 2000b) considers risk levels between 10^{-4} (one excess case of cancer for every 10 000 persons) and 10^{-6} (one excess case of cancer for every 1 000 000 persons) to be acceptable for the purpose of issuing fish and shellfish consumption advisories. Acceptable risk levels of 1×10^{-5} or 1×10^{-6} are most commonly used. However, because of the health benefits of consuming fish and shellfish some jurisdictions consider a risk of 1×10^{-4} as acceptable. Risks above 1×10^{-4} are usually considered unacceptable. For this risk assessment the acceptable risk level was defined as 1×10^{-4} .

According to the latest data³ the age standardised incidence of cancer amongst South African males is 129.41 per 100 000 and for females is 108.53 per 100 000. At a risk level of 1×10^{-4} this means a possibility of 10 additional incidences of cancer per 100 000.

To estimate the cumulative cancer risk due to simultaneous exposure to multiple chemicals in mussel and rock lobster tissue the Excess Cancer Risk for each chemical was summed to calculate a Total Cancer Risk.

6.4.6. Meal Limits

As discussed below, Excess and Total Cancer Risks

for high-end consumers exceeded an acceptable risk level of 1×10^{-4} . This does not mean mussels and rock lobsters cannot be consumed but that care should be taken in the number of meals consumed. The number of meals that can safely be consumed per month was calculated using a meal size of 140 g.

6.3. Results and Discussion

6.3.1. Morphological Measurements

The average total length and tail length of mussels and rock lobsters analysed is provided in Figure 6.3. The average total length of mussels was statistically similar between sites (One-Way ANOVA, $p = 0.058$). Size, which is presumed to reflect age, is thus unlikely to have been a significant factor influencing chemical concentration variability in the tissue of mussels at different sites. Although rock lobsters from different sites had statistically similar (and broadly comparable) average tail (Figure 6.3) and carapace lengths (ANOVA on Ranks, $p > 0.05$ in both cases), their whole wet weight varied fairly widely, but not statistically significantly between sites (ANOVA on Ranks, $p > 0.05$). Despite the lack of a statistically significant difference in rock lobster whole wet weight, which is partly driven by the small sample size, size may have had an influence on chemical concentration variability in the tissue of rock lobsters at different sites. However, for the purposes of this study rock lobster size was not considered a significant factor influencing chemical concentration variability.

6.3.2. Chemical Concentrations in Mussel and Rock Lobster Tissue

6.3.2.1. Metals

All metals apart from selenium were at a concentration exceeding the method detection limit in the tissue of mussels collected at each site on the Cape Peninsula in 2016 (Figure 6.4). Selenium was only detected in the tissue of mussels collected at Site M5 at Llandudno and Site M7 in Hout Bay Harbour. Ten of the 12 metals were at a concentration exceeding the method detection limit in the tissue of rock lobsters (Figure 6.4). Cadmium and lead were detected in the tissue of rock lobsters collected at three of the seven sites and in store purchased rock lobsters. Nickel and

³ [http://www.nioh.ac.za/assets/files/NCR_2011%20cancer%20tables%20\(2\)\(1\).pdf](http://www.nioh.ac.za/assets/files/NCR_2011%20cancer%20tables%20(2)(1).pdf)

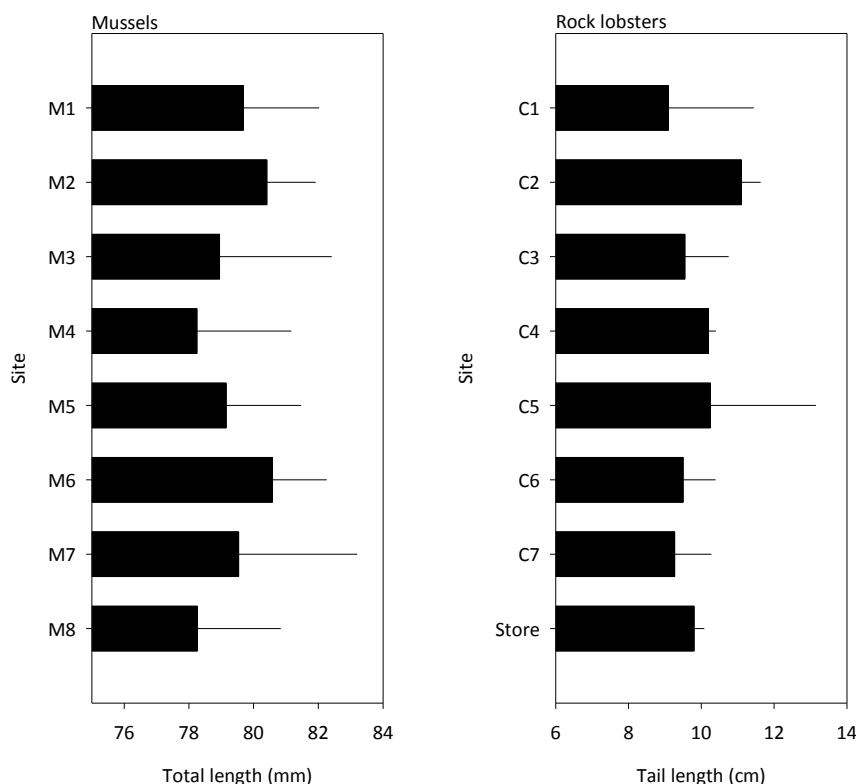


Figure 6.3. Average ($\pm$ standard deviation) total length of black mussels (*Mytilus galloprovincialis*) and tail length of west coast rock lobster (*Jasus lalandii*) collected at various sites along the Cape Peninsula area of Cape Town in 2016, and rock lobster purchased in a store.

selenium were never at a concentration exceeding the method detection limit in the tissue of rock lobsters (Figure 6.4).

Certain metals, such as copper, are essential for normal cellular functioning in most organisms, but at trace concentrations. However, even metals with no known physiological function (*e.g.* mercury) occur naturally in biological material, albeit usually at low concentrations (some fish and sharks naturally accumulate mercury to a relatively high concentration in their tissue). The presence of metals in biological tissue reflects the fact that essential and non-essential metals occur naturally in (sea)water, sediment and food/prey. The mere presence of metals in the tissue of mussels and rock lobsters collected along the Cape Peninsula does not, therefore, imply they were necessarily exposed to metal contaminated seawater and prey/food. In the context of this survey it is necessary to try and discriminate between metal concentrations in the tissue of mussels and rock lobsters that reflect exposure to natural metal concentrations and those that reflect bioaccumulation as a result of exposure to metal contaminated seawater, sediment and prey/food. This is not easy as metal concentrations

in the tissue of mussels and rock lobsters collected at a site at different times, and at different sites at the same time will invariably differ in the absence of exposure to metal contaminated seawater, sediment and prey/food. This reflects the differential bioaccumulation of metals (and other chemicals) as a function of mussel and rock lobster size (age) and sex, amongst other factors. The influence of size was, to the extent possible, accounted for by analysing mussels and rock lobsters of a broadly comparable size. Metal concentrations may also vary seasonally (*e.g.* due to upwelling). The simplest approach for determining if metal concentrations reflect bioaccumulation as a result of direct (water) or indirect (food) exposure to effluent constituents is thus to compare concentrations in mussels and rock lobsters collected inshore of the outfalls to concentrations in mussels and rock lobsters collected further afield. The hypothesis is mussels and rock lobsters inshore of the outfalls are more likely to be exposed to effluent constituents than those further afield, and if they are bioaccumulating effluent-derived metals then this should reflect in higher concentrations in their tissue (particularly metals at a high concentration in effluent). Mussels collected

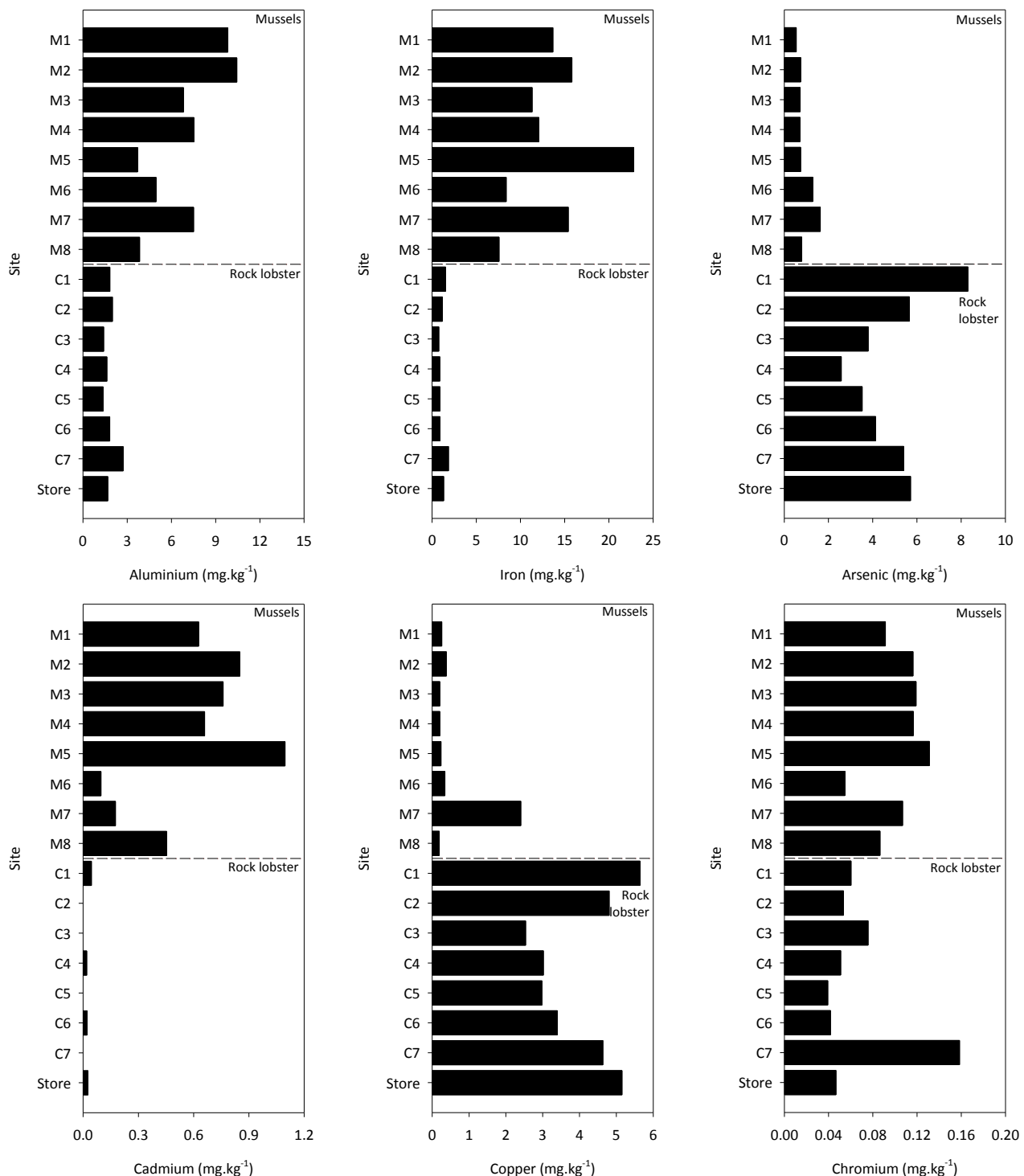


Figure 6.4. Metal concentrations (wet weight) in the tissue of black mussels (*Mytilus galloprovincialis*) and west coast rock lobsters (*Jasus lalandii*) collected at various sites along the Cape Peninsula area of Cape Town in 2016, and in rock lobsters purchased in a store.

at Site M2 at Sea Point, Site M5 at Llandudno, and Site M8 at Kommetjie, and rock lobsters collected at Site C2 at Sea Point and those purchased in a store are considered to have been collected sufficiently far from the outfalls to reflect baseline conditions.

There is no clear evidence mussels or rock lobsters collected inshore of the Green Point, Camps Bay

and Hout Bay outfalls had anomalously higher metal concentrations in their tissue than mussels collected at Sea Point, Llandudno and Kommetjie, and rock lobsters collected at Sea Point and purchased in a store (Figure 6.4). This conclusion is made in spite of the often considerable variability in the concentration of some metals between mussels and rock lobsters collected at different sites, and provided mussels collected at Site M7 in

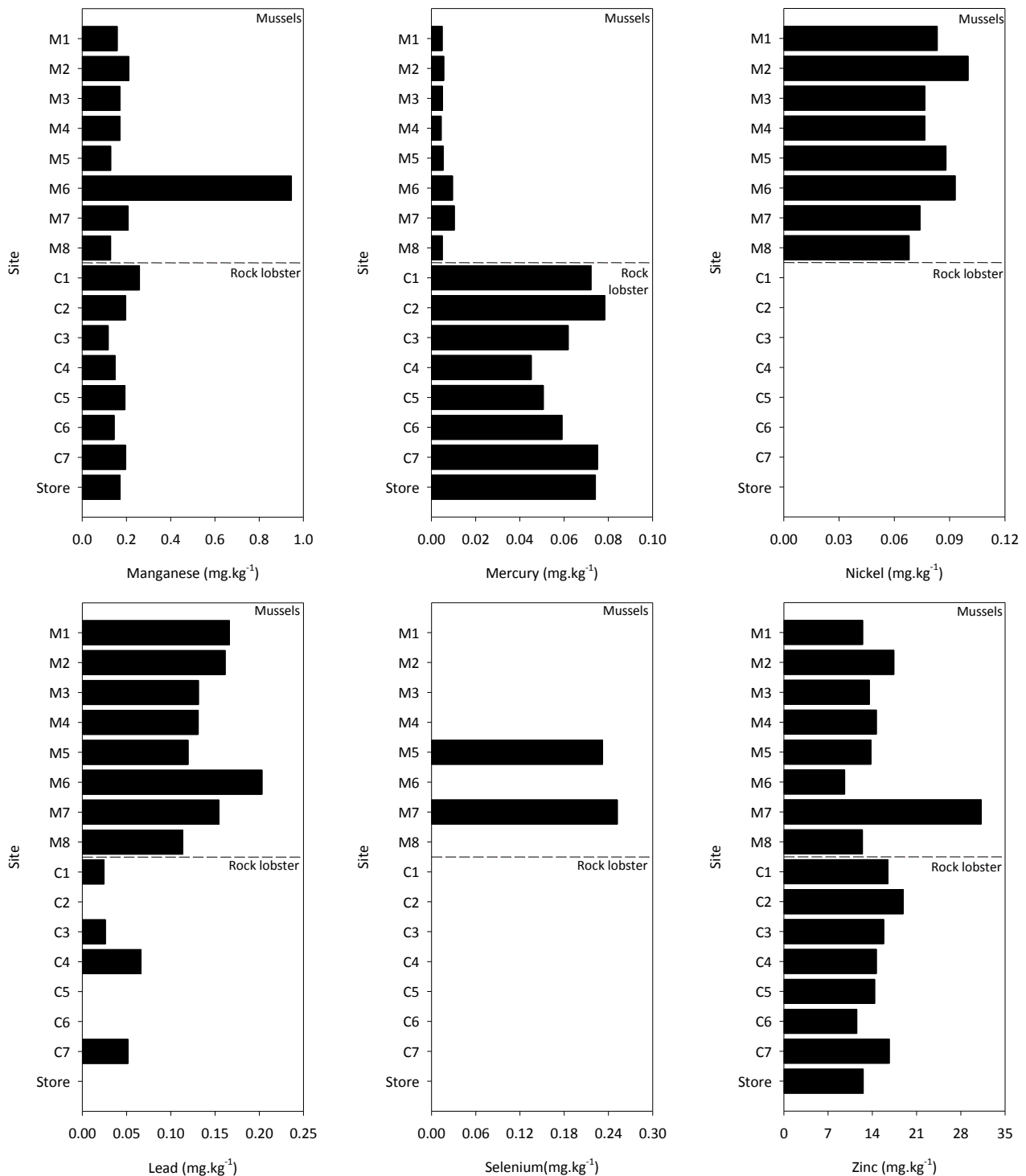


Figure 6.4 continued. Metal concentrations (wet weight) in the tissue of black mussels (*Mytilus galloprovincialis*) and west coast rock lobsters (*Jasus lalandii*) collected at various sites along the Cape Peninsula area of Cape Town in 2016, and in rock lobsters purchased in a store.

Hout Bay Harbour are ignored (see reason below). This is not to say concentrations of some metals were not highest in the tissue of mussels and rock lobsters collected inshore of certain of the outfalls, but there was no consistency across the different metals. The highest arsenic and copper concentrations, for example, were in the tissue of rock lobsters collected at Site C1 inshore of Green Point outfall. However, these were not inordinately

higher than concentrations in rock lobsters collected at Site C2 at Sea Point and those purchased in a store. Also, mercury and zinc concentrations in the tissue of rock lobsters collected at Site C2 at Sea Point were slightly higher than in rock lobsters collected at Site C1 inshore of the Green Point outfall. Similar trends are evident for mussels.

The lack of compelling evidence for the excessive bioaccumulation of effluent-derived metals does not mean at least a proportion of the concentration of some metals in the tissue of mussels and rock lobsters at some sites was not derived from effluent, but it is impossible to discriminate such source specific bioaccumulation with the data at hand. The question then is whether metal concentrations in mussels collected at all sites reflect bioaccumulation as a result of exposure to effluent. As discussed below the comparison of metal concentrations in the tissue of mussels collected for the 2015/2016 survey of the Cape Town outfalls monitoring programme to concentrations in mussels collected along the Cape Peninsula in other studies suggests this is not the case.

Apparent in Figure 6.4 is that mussels and rock lobsters had very different concentrations of certain metals in their tissue. Thus, aluminium, iron, cadmium and nickel were at a considerably higher concentration in the tissue of mussels while arsenic, copper and mercury were at a considerably higher concentration in the tissue of rock lobsters. For some metals this is explained by a difference in mussel and rock lobster physiology. The markedly higher concentration of copper in the tissue of rock lobsters, for example, is due to copper being an important constituent of haemocyanin in their (and other crustacean) haemolymph (blood) and being bound to enzymes in their muscle tissue (Bryan, 1968; Depledge and Bjerregard, 1998; Devescovi and Lucu, 2003). In other cases, such as zinc, mussels and rock lobster may have a different propensity for accumulating these metals in their tissue. Mussels and oysters, for example, show a difference in metal concentrations in their tissue in the absence of contamination and this is related to differences in their propensity for bioaccumulating the metals (e.g. Kimbrough *et al.*, 2008). The differences should not, therefore, automatically be assumed to reflect that mussels were exposed to certain contaminant metals more so than rock lobsters, and vice versa.

The cadmium concentration in the tissue of mussels collected at Site M1 at Mouille Point, Site M2 at Sea Point, Sites M3 and M4 at Camps Bay, and Site M5 at Llandudno is high compared to the concentration

in mussels (*Perna perna*) collected in the eThekweni area of KwaZulu-Natal (CSIR, unpublished data). The highest concentration was in mussels collected at Site M5 at Llandudno, which is relatively remote from the points of effluent discharge. The high concentration in mussels at Sites M1 - M5 might be explained by upwelling, an oceanographic feature that drives the high biological productivity in the marine environment along the west coast of South Africa. The cadmium concentration in seawater in coastal areas where upwelling is a prevalent are higher than in areas where upwelling is not prevalent (e.g. Bruland *et al.* 1978, Lares and Oriens 1997, Kavun *et al.* 2002, Cassis *et al.* 2011). Some microalgae (e.g. diatoms) are known to sequester cadmium in their frustules and this provides a potential exposure pathway considering mussels are filter feeders and phytoplankton accounts for about 30 - 40% of the nitrogen uptake in black mussels (Bustamante and Branch, 1996). However, studies have revealed that uptake across the gills and other exposed body surfaces directly from seawater is a far more significant route of exposure (Roesijadi and Robinson, 1994; Ettajani *et al.*, 2001; Zhanqiang, 2006; Strady *et al.*, 2011). The bioaccumulation of cadmium by cultured mussels and oysters in upwelling regions in Chile (Hervé-Fernández *et al.*, 2010), United States of America (Pacific Shellfish Institute, 2008) and Canada (Kruzynski, 2004) is so significant this has affected cross-border trade since concentrations in their tissue exceed health based limits. There is little evidence for cadmium contamination of seawater in these areas and the conclusion is the bioaccumulation reflects exposure to cadmium in upwelled water. Cadmium has also been detected in the tissue of mussels cultured in Saldanha Bay at concentrations that exceed the limit ($1 \mu\text{g.g}^{-1}$ wet weight) for trade with the European Union. Although some investigators (e.g. Clarke *et al.*, 2015) contend the high concentrations reflect bioaccumulation as a result of exposure to contaminated seawater and food, the source may well be upwelled water. If upwelling does account for the high cadmium concentration in mussels along the Cape Peninsula then it is interesting that mussels at Site M6 in Hout Bay, Site M7 in Hout Bay Harbour, and Site M8 in Kommetjie had lower concentrations than at other sites.

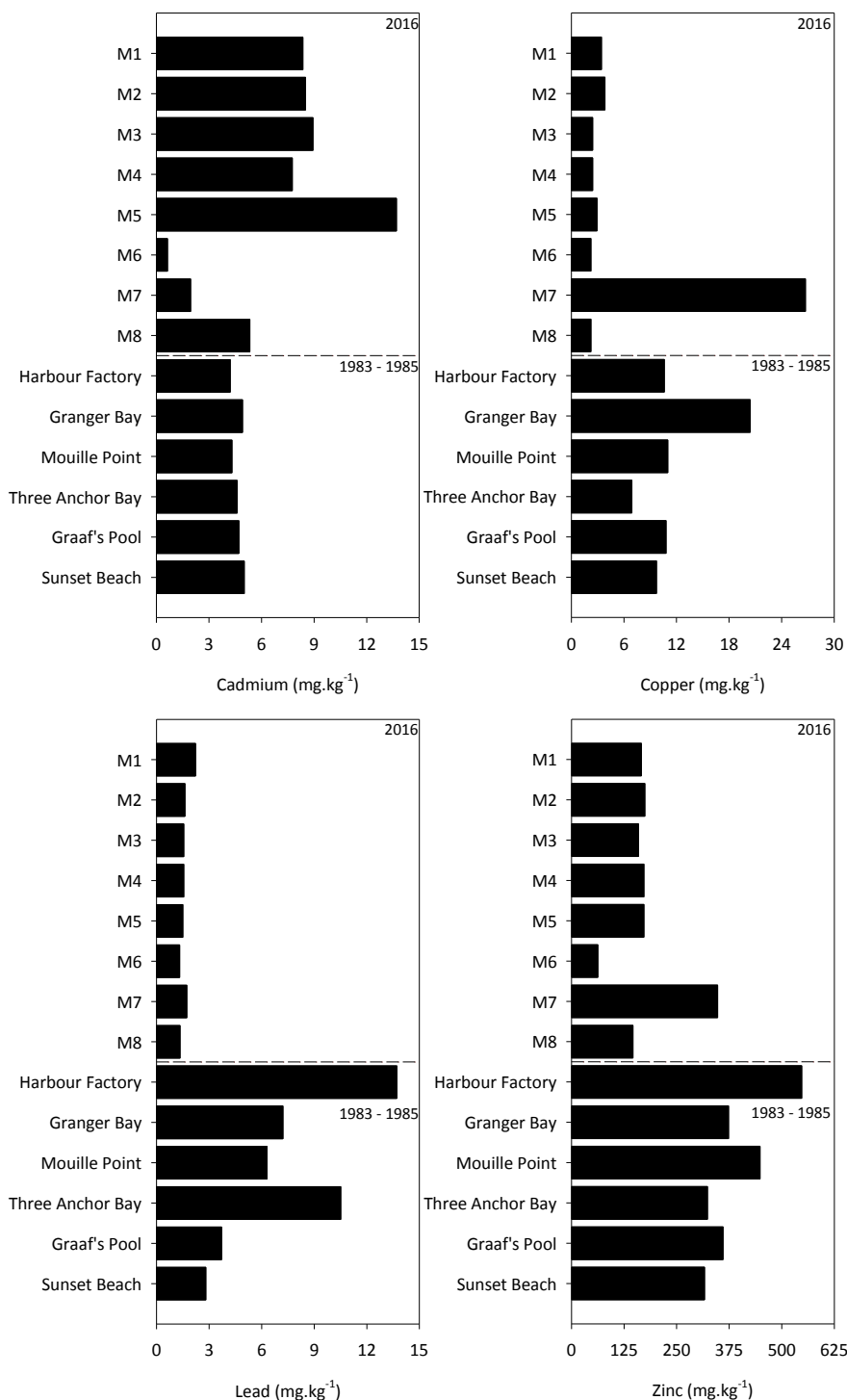


Figure 6.5. Comparison of metal concentrations (dry weight) in the tissue of black mussels (*Mytilus galloprovincialis*) collected at various sites along the Cape Peninsula area of Cape Town in 2016 to concentrations in black mussels collected between Harbour Factory and Sunset Beach in 1983 - 1985 (Steering Committee, 1985).

The concentration of copper and zinc in the tissue of mussels collected at Site M7 in Hout Bay Harbour was far higher than in mussels at other sites. This almost certainly reflects exposure of mussels in Hout Bay Harbour to copper and zinc derived from anthropogenic sources other than effluent. As discussed in Chapter 5, copper and zinc are important biocides in antifouling coatings on the hulls of vessels and are amongst the most common

and significant contaminants of sediment in ports. The reader might recall from Chapter 5 that sediment at two sites in Hout Bay Harbour was the, or amongst the most significantly contaminated by copper and zinc for sediment collected at any site for the 2015/2016 survey of the Cape Town outfalls monitoring programme.

Figures 6.5 and 6.6 compare metal concentrations

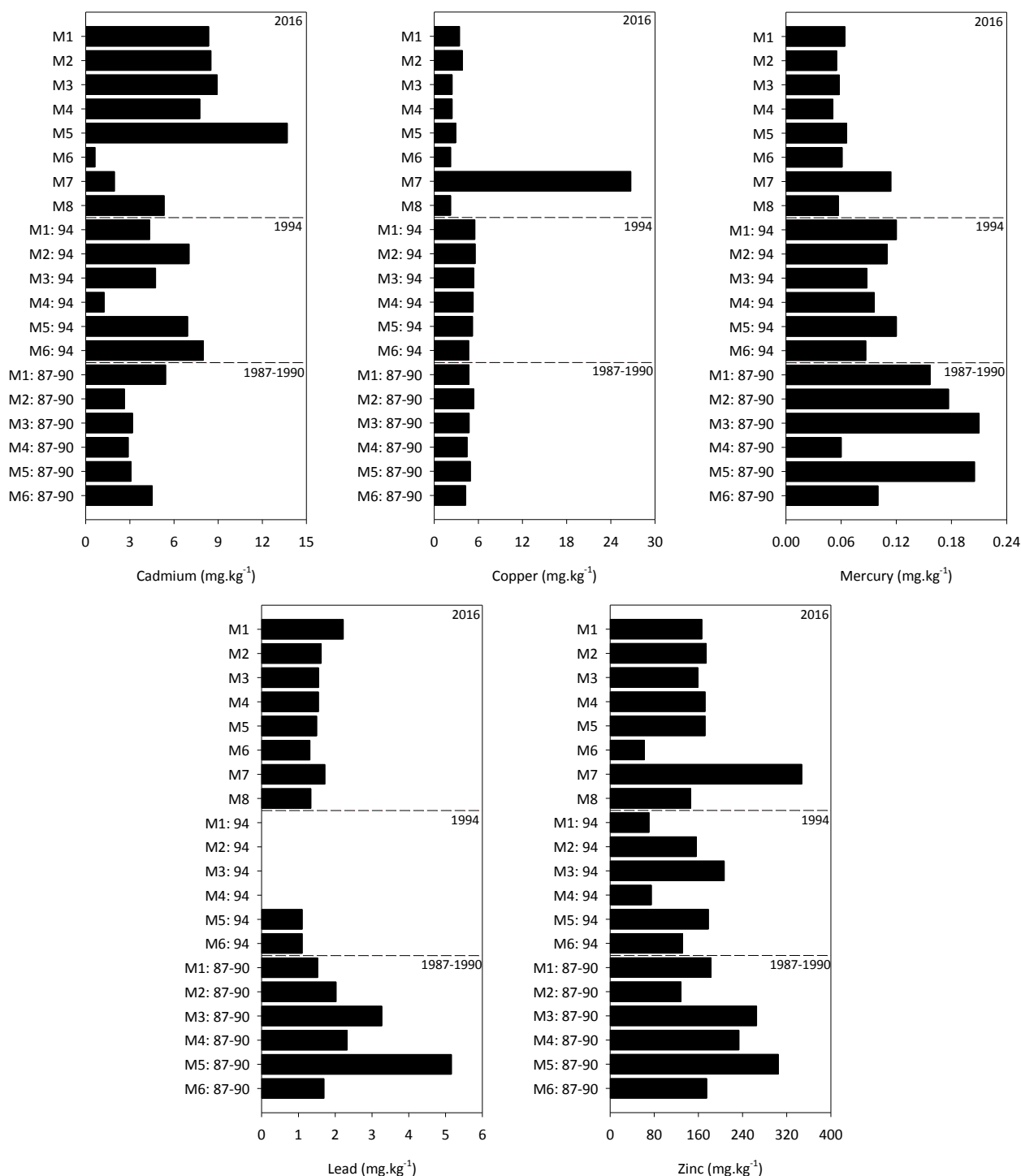


Figure 6.6. Comparison of metal concentrations (dry weight) in the tissue of black mussels (*Mytilus galloprovincialis*) collected at various sites along the Cape Peninsula area of Cape Town in 2016 to concentrations in black mussels collected in Hout Bay between 1987 - 1990 (CSIR, 1993) and in 1994 (CSIR, 1995). Where no data are indicated the metal was at a concentration below the method detection limit. The bars for the period 1987 - 1990 represent the mean of measurements made on 6 - 7 samples of mussels collected over this period (standard deviations are not shown for display purposes).

in the tissue of black mussels collected for 2015/2016 survey of the Cape Town outfalls monitoring programme to average concentrations in the tissue of black mussels collected at various sites between Granger Bay and Sunset Beach in 1983 - 1985 (Steering Committee, 1985) and at six sites in Hout Bay between 1987 - 1990 (CSIR, 1993) prior to the

commissioning of the outfall and in 1994 after the commissioning of the outfall (CSIR, 1995). The reader should note the average concentration for mussels collected between 1983 - 1985 and 1987 - 1990 sometimes masks a fairly significant (annual) variation in the concentration of some metals at some sites. Also, the methods used to extract and

analyse metal concentrations in mussels collected for these surveys were not identical to methods used in this study.

Copper, lead and zinc concentrations in mussels collected between Granger Bay and Sunset Beach in 1983 - 1985 were higher than in mussels collected along the Cape Peninsula in 2016 apart from those collected at Site M7 in Hout Bay Harbour (Figure 6.5). Cadmium concentrations in mussels collected at Sites M1 - M5 in 2016 were, in turn, higher than concentrations in mussels collected between 1983 - 1985. Although this suggests copper, lead and zinc concentrations in mussels have declined with time it is uncertain if the difference is real or the result of different methods used to analyse metal concentrations in mussels between the surveys.

Metal concentrations in mussels collected in Hout Bay in 1994 were generally comparable to concentrations in mussels collected on the Cape Peninsula in 2016, with two notable exceptions (Figure 6.6). First, lead was not detected in mussels collected at four of the six sites in 1994. Second, cadmium concentrations in mussels collected at Sites M1 - M5 in 2016, but not in mussels collected at Sites M6 and M7 in Hout Bay and Site M8 at Kommetjie, are higher than in mussels collected in 1994. Concentrations of most metals in mussels collected in Hout Bay between 1987 - 1990 are broadly comparable to concentrations in mussels collected in 2016 (and in 1994 where applicable), but lead and zinc concentrations tended to be somewhat higher between 1987 - 1990. These comparisons provide no evidence mussels collected along the Cape Peninsula in 2016 had significantly higher concentrations of metals in their tissue than mussels collected in Hout Bay between 1987 - 1994. The data in fact suggest a slight decrease in the concentration of several metals over time, but this might again reflect a difference in methods used to analyse metal concentrations in mussels between the surveys.

Sparks *et al.* (2014) provide a summary of metal concentrations in black mussels collected for a Mussel Watch Programme run by the Department of Environmental Affairs between 1985 - 2008. Black mussels were collected at five sites, namely Site 1 comprising subsites at Olifantsbos and

Noordhoek, Site 2 comprising five subsites in Hout Bay, Site 3 comprising three subsites in Sea Point, Three Anchor Bay and Granger Bay, Site 4 comprising four subsites in Table Bay, and Site 5 comprising three subsites in Milnerton, Paarden Island and Bloubergstrand. Concentrations of all metals apart from cadmium in mussels collected along the Cape Peninsula in 2016 are comparable or slightly lower than the average concentration in mussels collected between 1985 - 2008. The cadmium concentration in mussels collected at sites M1 - M5 in 2016 was in turn higher than the average concentration in mussels collected between 1985 - 2008.

Metal concentrations in mussels collected along the Cape Peninsula in 2016 are within the range reported for mussels in relatively uncontaminated parts of the Mediterranean Sea with the exception of cadmium, which was often at a considerably higher concentration in mussels along the Cape Peninsula (*e.g.* Çullaj *et al.*, 2006; Desideri *et al.*, 2010; Ramsak *et al.*, 2012; Spada *et al.*, 2013; Jović and Stanković, 2014).

As far as the scientists that prepared this report could establish there is no published information on metal concentrations in the tissue of west coast rock lobster *Jasus lalandii*. No comparison to the findings of other studies is thus possible.

6.3.2.2. Polycyclic Aromatic Hydrocarbons

Polycyclic aromatic hydrocarbons were at a concentration exceeding the method detection limit in the tissue of mussels collected at all sites along the Cape Peninsula in 2016 (Figures 6.7 and 6.8). Seven isomers were detected in mussels at Site M1 at Mouille Point and eight in mussels at Site M6 near Hout Bay Harbour and Site M7 in the harbour. At other sites one or two isomers were detected in mussels. By far the highest total polycyclic aromatic hydrocarbon concentration was in mussels at Site M7 in Hout Bay Harbour ($15.58 \mu\text{g.kg}^{-1}$ wet weight) followed by mussels at Site M6 near the harbour ($6.22 \mu\text{g.kg}^{-1}$ wet weight) (Figures 6.7 and 6.8). Total polycyclic aromatic hydrocarbon concentrations in the tissue of mussels at other sites were broadly comparable ($0.42 - 1.21 \mu\text{g.kg}^{-1}$ wet weight). The high concentration in mussels at Sites M6 and M7, and the isometric composition

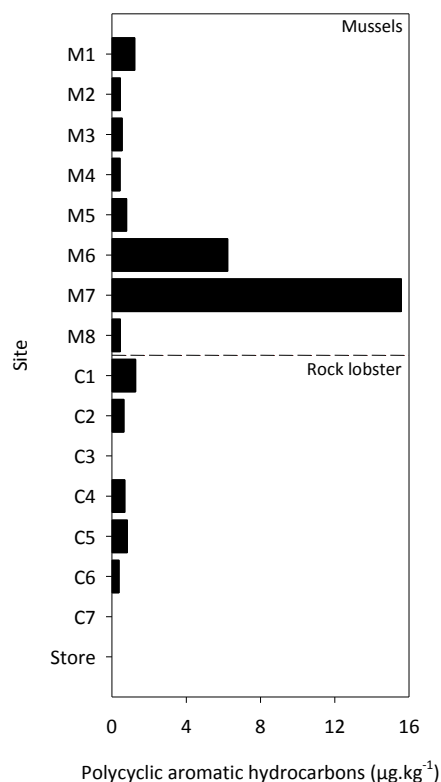


Figure 6.7. Total polycyclic aromatic hydrocarbon concentrations (wet weight) in the tissue of black mussels (*Mytilus galloprovincialis*) and west coast rock lobsters (*Jasus lalandii*) collected at various sites along the Cape Peninsula area of Cape Town in 2016.

undoubtedly reflect exposure of the mussels to polycyclic aromatic hydrocarbons from a local source. For mussels at Site M7 this probably reflects, at least in part, exposure to oil and lubricants spilled from vessels. Mussels at Site M1 at Mouille Point, inshore of the Green Point outfall, had a slightly higher total polycyclic aromatic hydrocarbon concentration than mussels collected at most other sites. Although this might reflect bioaccumulation as a result of exposure to effluent-derived polycyclic aromatic hydrocarbons there is no evidence mussels at Site M3 inshore of (and very near to) the Camps Bay outfall, and at Site M4 in Balie Bay near Camps Bay had excessively accumulated these chemicals in their tissue. In fact, the total concentration in mussels at Site M5 in Llandudno, distant from effluent discharge points, was higher than in mussels at Sites M3 and M4. The isomer composition of polycyclic aromatic hydrocarbons in mussels at Site M1 suggests, furthermore, the mussels were exposed to a localised source/s of these chemicals that are distinct from effluent.

Polycyclic aromatic hydrocarbons were at a concentration exceeding the method detection limit in the tissue of rock lobsters collected at five of the seven sites on the Cape Peninsula in 2016, but only two isomers were detected (Figures 6.7 and 6.9). Interestingly, one of the isomers, namely cyclopenta(c,d)pyrene, was never detected in the tissue of mussels. This suggests a pathway favouring cyclopenta(c,d)pyrene accumulation in rock lobsters but not mussels. Polycyclic aromatic hydrocarbons were not detected in rock lobsters purchased in a store. The total polycyclic aromatic hydrocarbon concentration in the tissue of rock lobsters was broadly comparable between sites (0.37 - 1.26 µg.kg⁻¹ wet weight) and to concentrations in mussels apart from those at Sites M6 and M7 in and near Hout Bay Harbour.

According to system for classifying contaminant concentrations in mussels in Norway (Molvær *et al.*, 1997; Knutzen and Green, 2001) the total polycyclic aromatic hydrocarbon concentration in mussels collected at all sites along the Cape Peninsula in 2016 (highest concentration = 15.58 µg.kg⁻¹ wet weight) are insignificant (<50 µg.kg⁻¹ wet weight). Total polycyclic aromatic hydrocarbon concentrations (all dry weight) in the same species of mussel (*M. galloprovincialis*) as that collected on the Cape Peninsula in 2016 from the Tohoku district in Japan were 129 - 181 µg.kg⁻¹ (Tanaka and Onduka, 2010) and from the Mediterranean coast in France and Spain were 17.9 - 58.9 µg.kg⁻¹ (Baumard *et al.*, 1998). Thus, the total polycyclic aromatic hydrocarbon concentration in mussels collected along the Cape Peninsula in 2016 were lower than concentrations in these studies apart from in mussels at Site M6 in Hout Bay (40.15 µg.kg⁻¹) and Site M7 in Hout Bay Harbour (173.08 µg.kg⁻¹).

Based on the spatial trend in total polycyclic aromatic hydrocarbons in the tissue of mussels and rock lobsters collected along the Cape Peninsula in 2016 there is no compelling evidence they had excessively accumulated these chemicals in their tissue as a result of exposure to effluent constituents. If this was the case the expectation would be for the concentration to be higher in mussels and rock lobsters collected inshore of the outfalls than in those further afield. This does not mean at least some of the polycyclic aromatic



Figure 6.8. Bubble plot representation of total polycyclic aromatic hydrocarbon concentrations in the tissue of black mussels (*Mytilus galloprovincialis*) collected at various sites along the Cape Peninsula area of Cape Town in 2016. The largest bubble represents a concentration of $15.58 \mu\text{g.kg}^{-1}$ (wet weight).

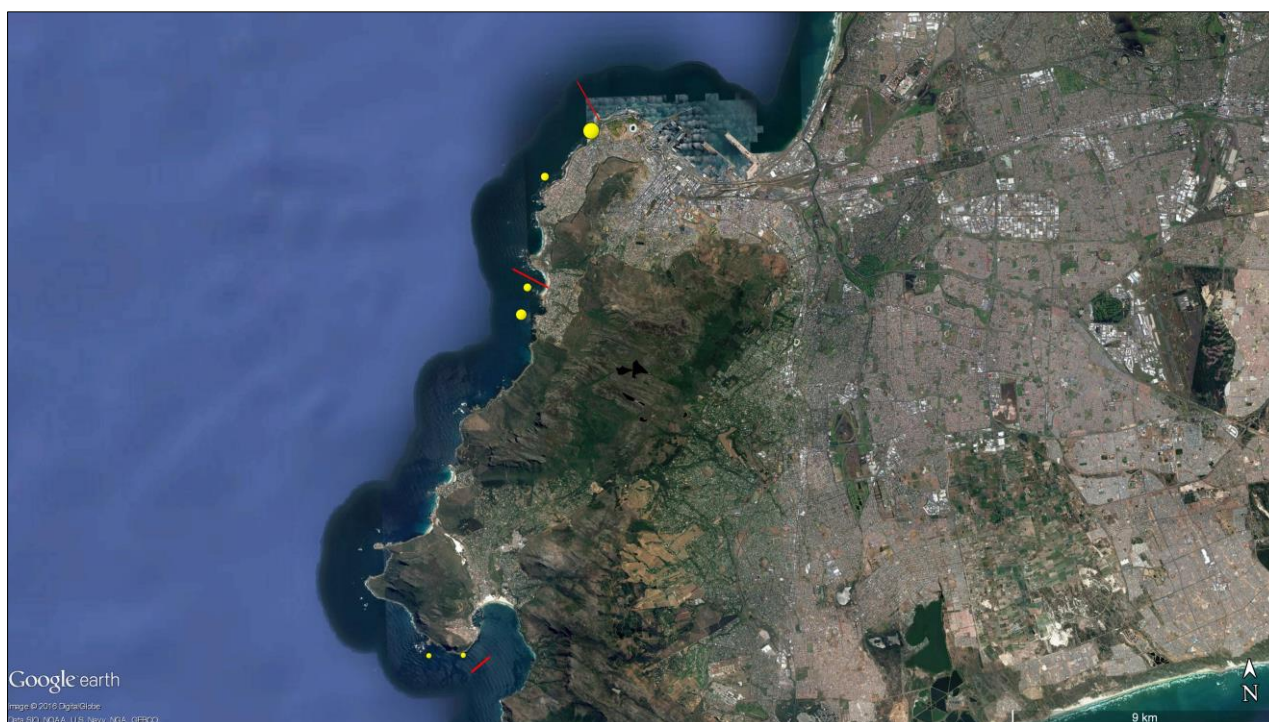


Figure 6.9. Bubble plot representation of total polycyclic aromatic hydrocarbon concentrations in the tissue of west coast rock lobsters (*Jasus lalandii*) collected at various sites along the Cape Peninsula area of Cape Town in 2016. The largest bubble represents a concentration of $1.27 \mu\text{g.kg}^{-1}$ (wet weight).

hydrocarbons in mussels and rock lobsters at some sites were not derived from effluent, but if so it is impossible to identify such source specific bioaccumulation with the data at hand.

Mason (1988) analysed hydrocarbon concentra-

tions in the tissue of three species of mussel collected along the Table Bay, Cape Peninsula and False Bay shorelines in 1985 and 1986. Two aromatic hydrocarbon fractions (2 and 3 ring, and 4 or more rings) were analysed by fluorescence, but Mason (1988) did not provide concentrations for

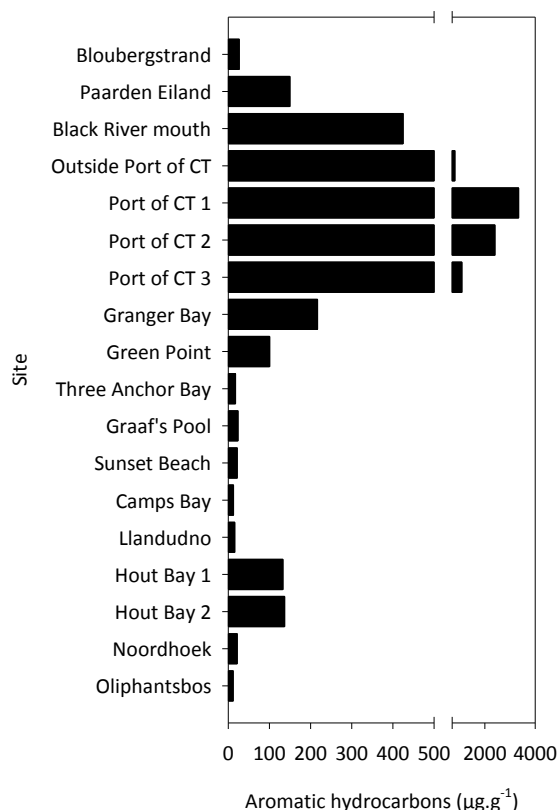


Figure 6.10. Hydrocarbon concentrations (dry weight) in the tissue of black mussels (*Mytilus galloprovincialis*) collected Between Bloubergstrand (BB) in Table Bay and Oliphantsbos (OB) on the Cape Peninsula in 1985 by Mason (1988).

individual isomers. The concentrations reported by Mason (1988) are orders of magnitude higher than those in the tissue of mussels collected in 2016. This presumably reflects a methodological difference. It thus makes little sense to directly compare the data save to state the spatial trend in concentration for black mussels collected at 17 sites between Bloubergstrand in Table Bay and Oliphantsbos on the Cape Peninsula, but not including mussels collected in the Port of Cape Town and in Hout Bay Harbour, is similar to the trend for polycyclic aromatic hydrocarbon concentrations in mussels collected in 2016 (Figure 6.10). Hydrocarbon concentrations in mussels collected in the Port of Cape Town were far higher than in mussels elsewhere. Concentrations in mussels at Paarden Island, at the mouth of the Black (Salt) River, and in and near Hout Bay Harbour were considerably higher than at most other sites.

As far as the scientists that prepared this report could establish there is no published information on polycyclic aromatic hydrocarbon concentrations in the tissue of west coast rock lobster *Jasus*

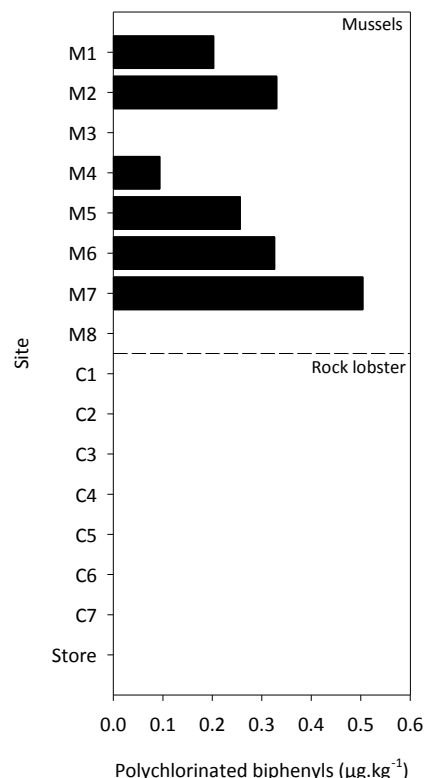


Figure 6.11. Total polychlorinated biphenyl concentration (wet weight) in the tissue of black mussels (*Mytilus galloprovincialis*) and west coast rock lobsters (*Jasus lalandii*) collected at various sites along the Cape Peninsula area of Cape Town in 2016.

lalandii. No comparison to the findings of other studies is thus possible.

6.3.2.3. Polychlorinated Biphenyls

Polychlorinated biphenyls were at a concentration exceeding the method detection limit in the tissue of mussels collected at six of the eight sites on Cape Peninsula in 2016, but not in the tissue of rock lobsters collected in the field or purchased in a store (Figures 6.11 and 6.12). However, only PCB's 137 and 153 were detected. These are amongst the most recalcitrant of congeners and often comprise a significant proportion of the total polychlorinated biphenyl concentration in marine fish and shellfish. The highest total concentration was in mussels collected at Site M7 in Hout Bay Harbour, but this was not markedly higher than the concentration in mussels at Site M2 at Sea Point, Site M5 at Llandudno, and Site M6 near Hout Bay Harbour. According to system for classifying contaminant concentrations in mussels in Norway (Molvær *et al.*, 1997; Knutzen and Green, 2001) the total polychlorinated biphenyl concentration in all rock lobsters (highest concentration = 0.504 µg.kg⁻¹ wet



Figure 6.12. Bubble plot representation of polychlorinated biphenyl concentrations (wet weight) in the tissue of black mussels (*Mytilus galloprovincialis*) collected at various sites along the Cape Peninsula area of Cape Town in 2016.

weight) is insignificant ($<3 \mu\text{g.kg}^{-1}$ wet weight).

As stated above, polychlorinated biphenyls were not detected in four effluent samples collected from the Green Point, Camps Bay and Hout Bay wastewater works in 2016 and were considered a possible tracer of contaminants introduced to the marine environment off Cape Town from land-based sources other than effluent. If correct the implication is at least some of the polycyclic aromatic hydrocarbon and possibly also metal concentrations in the tissue of mussels collected at most sites along the Cape Peninsula in 2016 were derived from land-based sources other than effluent.

As far as the scientists that prepared this report could establish there are no published studies on polychlorinated biphenyl concentrations in the tissue of west coast rock lobster *Jasus lalandii*. No comparison to the findings of other studies is thus possible.

6.3.2.4. Non-Steroidal Anti-Inflammatory Drugs

Non-steroidal anti-inflammatory drugs were not detected in the tissue of mussels or rock lobsters collected along the Cape Peninsula in 2016. This suggests mussels and rock lobsters are either not

bioaccumulating these compounds in their tissue due to their properties (*e.g.* low lipophilicity; but see McEneff *et al.* [2014] for evidence of non-steroidal anti-inflammatory drug bioaccumulation by mussels) or they are not (persistently reaching) the nearshore and shoreline at high concentrations. It is impossible with the data at hand to determine which of these possible explanations is more likely.

6.3.3. Risk Assessment

6.3.3.1. Non-Carcinogenic Risk

The Hazard Quotient for each chemical analysed in mussel and rock lobster tissue does not exceed a value of one for low- and high-end consumers apart from cadmium in mussels collected at Site M5 in Llandudno for high-end consumers (Figures 6.13 and 6.14). The Hazard Quotient for cadmium in these mussels was 1.17. Mussels and rock lobsters collected along the Cape Peninsula can thus be consumed with little risk of systemic health effects as a result of exposure to chemicals in their tissue. This conclusion is made in spite of the Hazard Quotient for cadmium in mussels at Site M5 exceeding a value of one, since the quotient only slightly exceeds a value of one and there is a substantial safety factor included in the Reference Dose.

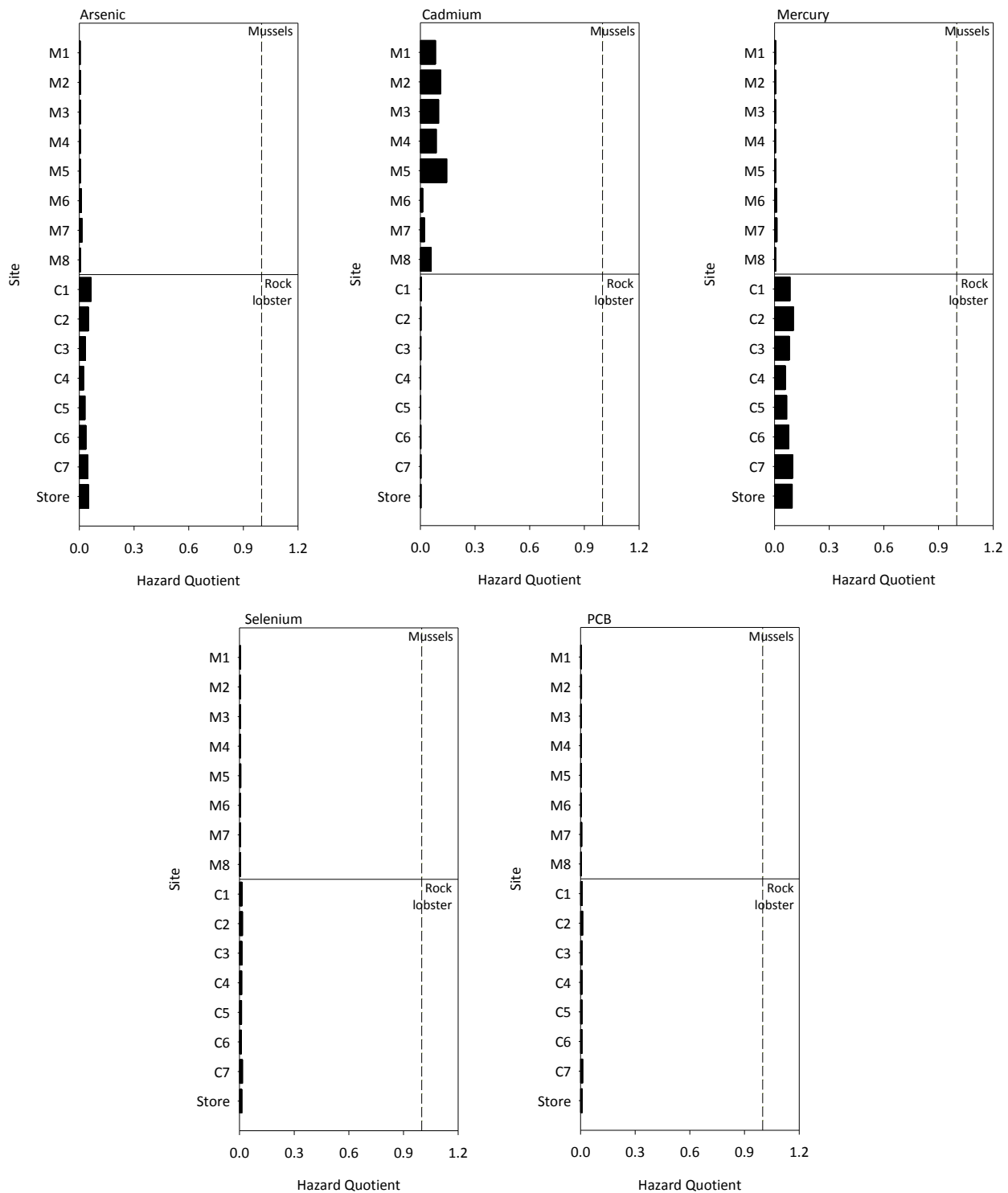


Figure 6.13. Hazard Quotients for various chemicals in black mussels (*Mytilus galloprovincialis*) and west coast rock lobster (*Jasus lalandii*) collected at sites along the Cape Peninsula area of Cape Town in 2016 as per the low-end consumer scenario. The vertical dashed lines represent a Hazard Quotient of one. The x-axis of the graphs is set to the same scale to highlight differences and similarities in quotients for different chemicals.

6.3.3.2. Carcinogenic Risk

The Excess Cancer Risk for inorganic arsenic, polycyclic aromatic hydrocarbons and polychlorinated biphenyls, and Total Cancer Risk for the combined effect of these chemicals for low-end consumers is below 1×10^{-4} for mussels and rock lobsters at all sites, and for store purchased rock lobsters (Figures

6.15 and 6.17).

The Excess Cancer Risk for polycyclic aromatic hydrocarbons and polychlorinated biphenyls for high-end consumers is below 1×10^{-5} (Figure 6.16). The risk posed by inorganic arsenic in rock lobsters collected at Sites C1, C2 and C7 and those

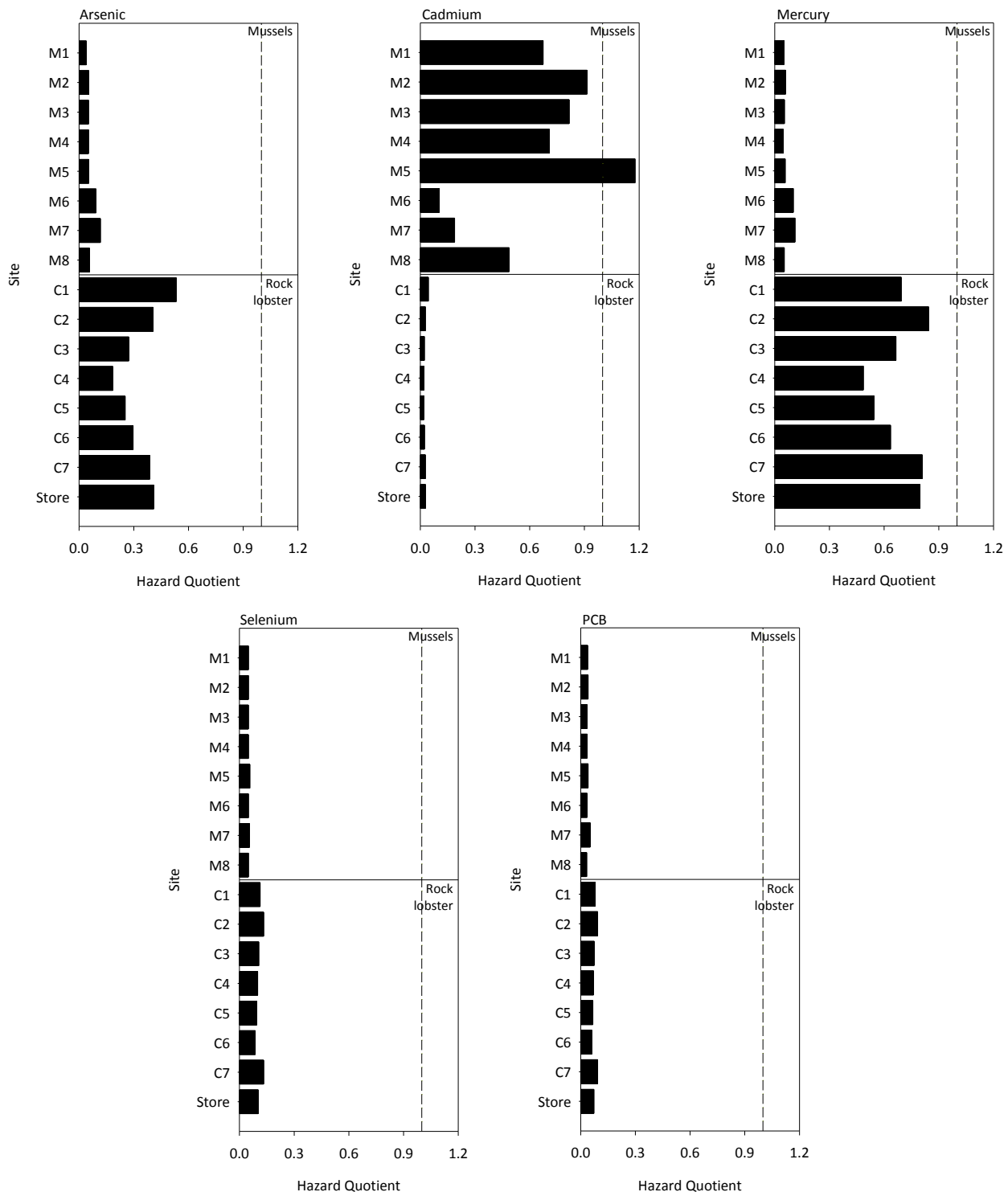


Figure 6.14. Hazard Quotients for various chemicals in black mussels (*Mytilus galloprovincialis*) and west coast rock lobsters (*Jasus lalandii*) collected at sites along the Cape Peninsula area of Cape Town in 2016 as per the high-end consumer scenario. The vertical dashed lines represent a Hazard Quotient of one. The x-axis of the graphs is set to the same scale to highlight differences and similarities in quotients for different chemicals.

purchased in a store slightly exceeds 1×10^{-4} . As a result the Total Cancer Risk also slightly exceeds 1×10^{-4} (Figure 6.17). The highest Total Cancer Risk was 1.76×10^{-4} for rock lobsters collected at Site C1 at Green Point followed by 1.36×10^{-4} for rock lobsters at Site C2 at Sea Point. In other words there is chance of about 1.8 and 1.3 additional

incidences of cancer per 10 000 members of the population consuming rock lobster from these sites per the high-end consumer scenario.

Inorganic arsenic is the most important contributor to the Total Cancer Risk for high-end consumers (93 - 97% for rock lobsters and 82 - 92% for

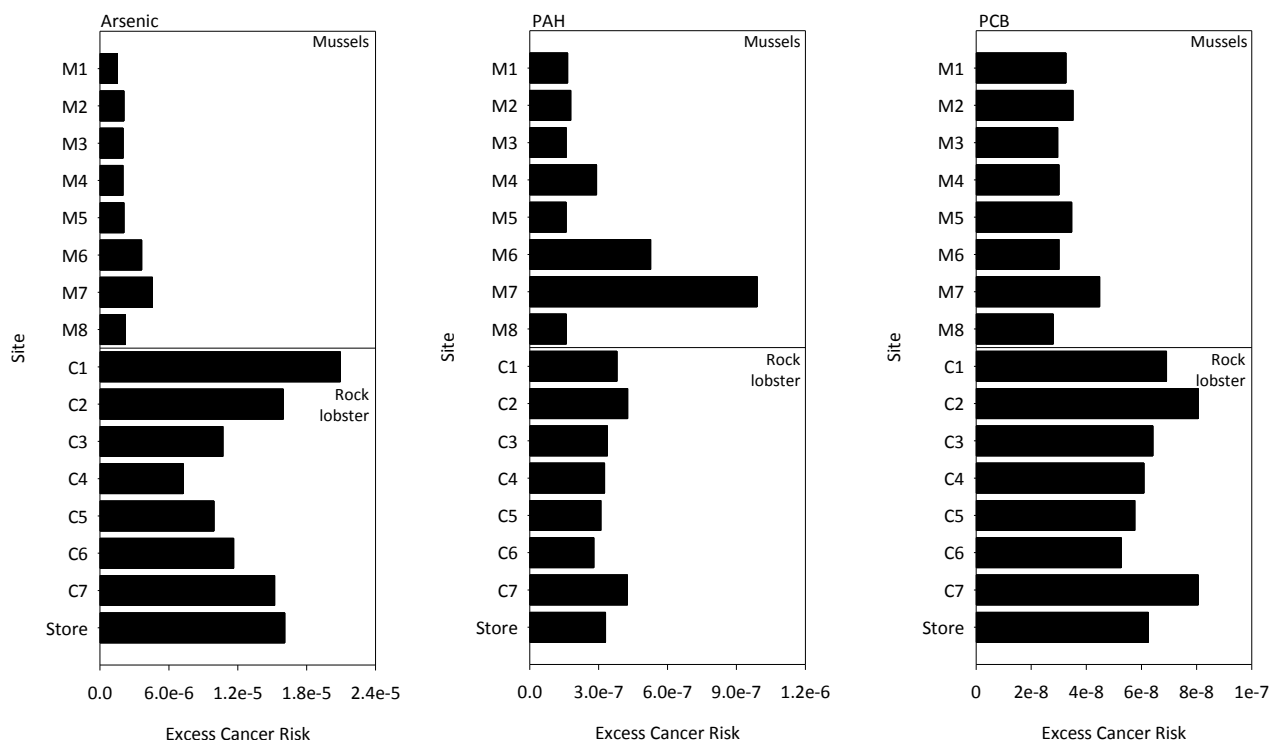


Figure 6.15. Excess Cancer Risk for various chemicals in black mussels (*Mytilus galloprovincialis*) and west coast rock lobsters (*Jasus lalandii*) collected at sites along the Cape Peninsula area of Cape Town in 2016 and rock lobsters purchased in a store as per the low-end consumer scenario.

mussels). Based on the contribution the question is if rock lobsters along the Cape Peninsula are accumulating arsenic in their tissue due to the widespread contamination of nearshore coastal waters by this metal. There is no evidence the arsenic was derived from effluent. Marine animals have a limited ability to bioaccumulate inorganic arsenic from seawater or food (Francesconi and Edmonds, 1997). Arsenic was also at a low concentration in effluent collected from the Green Point, Camps Bay and Hout Bay wastewater works between 1 March 2015 and 30 September 2016 (see Chapter 2). The arsenic concentration in effluent was in fact comparable to or lower than that expected naturally in seawater.

The arsenic concentration in west coast rock lobster can be placed into perspective by comparing this to the concentration in lobsters from other parts of the world. Lobsters of several genera, including *Homarus*, *Nephrops*, *Jasus* and *Thenus*, may contain more than 100 mg.kg⁻¹ and occasionally more than 200 mg.kg⁻¹ (dry weight) of arsenic in their abdominal muscle (Falconer *et al.*, 1983; Neff, 1997). Flanjak (1982) reported a mean total arsenic concentration in *Jasus verreauxii* from two areas near Sydney and purchased at the

Sydney Fish Market of 25 mg.kg⁻¹ (range = 12 - 54 mg.kg⁻¹, wet weight), with inorganic arsenic contributing about 0.01% of the total arsenic concentration. Fabris *et al.* (2006) report mean total arsenic concentrations in *Jasus edwardsii* from four areas in Victoria in Australia of 45.0 - 67.4 mg.kg⁻¹ (presumed dry weight), with inorganic arsenic contributing about ≤0.2% of the total arsenic concentration. Peshut *et al.* (2008) provide a median arsenic concentration in *Panulirus* sp. lobsters from American Samoa of 48.8 mg.kg⁻¹ (range = 19.8 - 99.2 mg.kg⁻¹, wet weight), with inorganic arsenic contributing about 0.05% (range <0.01 - 0.2%) of the total arsenic concentration. The average arsenic concentration in lobster (species not identified) reported by Australian food safety authorities for the period 2000 - 2013 is 45 mg.kg⁻¹ (range = 5.7 - 150 mg.kg⁻¹, wet weight), with inorganic arsenic contributing about 0.05% (range 0.008 - 0.11%) of the total arsenic concentration. Total arsenic concentrations analysed in west coast rock lobster *Jasus lalandii* collected on the Cape Peninsula in 2016, which ranged between 4.88 - 8.30 mg.kg⁻¹ (wet weight), are thus generally far lower than concentrations in lobsters in other parts of the world. Also, the contribution of inorganic arsenic to the total arsenic concentration is far

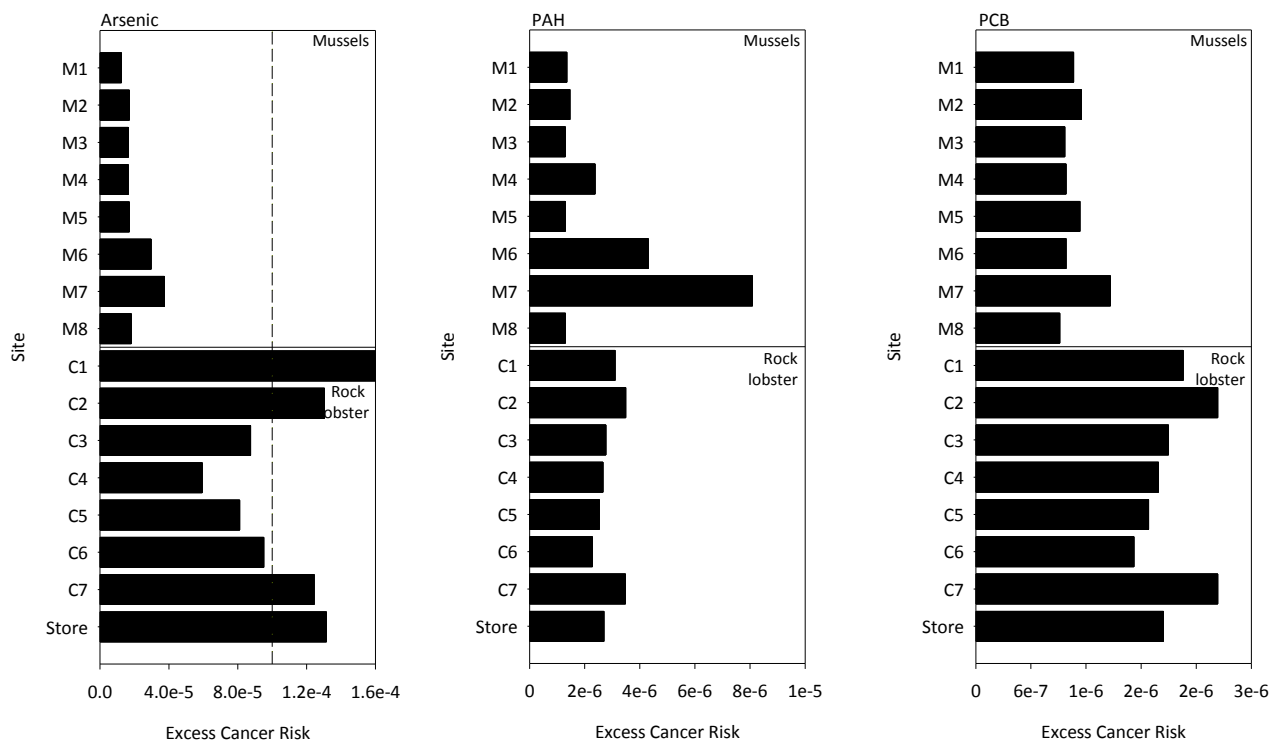


Figure 6.16. Excess Cancer Risk for various chemicals in black mussels (*Mytilus galloprovincialis*) and west coast rock lobster (*Jasus lalandii*) collected at sites along the Cape Peninsula area of Cape Town in 2016 and rock lobsters purchased in a store as per the high-end consumer scenario. The vertical dashed line represents a risk of 1×10^{-4} .

lower than that assumed for this study. Although the direct comparison of metal concentrations between species and genera should be treated with caution, the data provided by Fabris *et al.* (2006) suggest metal concentrations in west coast rock lobster are not anomalously high for *Jasus* spp.

If the contribution of inorganic arsenic in this risk assessment was assumed to be 0.2%, in line with that for other lobsters, the Total Cancer Risk for all rock lobsters collected along the Cape Peninsula in 2016 is below 1×10^{-5} . This contribution was not, however, considered in the absence of an empirical understanding on the ratio of inorganic to total arsenic in west coast rock lobster, in the interest of a conservative risk assessment.

6.3.3.3. Meal Consumption Limits

More than one 140 g meal of mussels collected at all sites on the Cape Peninsula in 2016 can be safely consumed per day (Figure 6.18). For rock lobsters between 7 - 20 meals can be consumed per month depending on the collection site (Figures 6.18). This is based on an assumed inorganic to total arsenic concentration ratio of 2%. If the ratio is assumed to be 0.2%, which is in line with measurements for other lobsters, then more than one 140 g meal can

be safely consumed per day.

6.3.3.4. Risk Assessment Uncertainties

Although this risk assessment identified possible carcinogenic risks to the health of humans that consume rock lobsters collected at some sites on the Cape Peninsula or those purchased in a store as per the high-end consumer scenario, it is important to understand the uncertainties associated with the risk assessment. These arise due to the lack of information on some variables required to calculate risk and the consequent need to make assumptions on the value of these variables.

First, risk was assessed based on exposure to chemical concentrations analysed in the tissue of black mussels and rock lobsters collected in 2016. The assumption is these will not change through the 30 year period used to assess chronic and carcinogenic risks to low- and high-end consumers. It is unlikely concentrations of organic chemicals will remain unchanged in mussels and rock lobsters for this period and at least some annual or inter-annual variation in metal concentrations can also be expected, even in the absence of contamination.

Second, only chemicals analysed for this survey

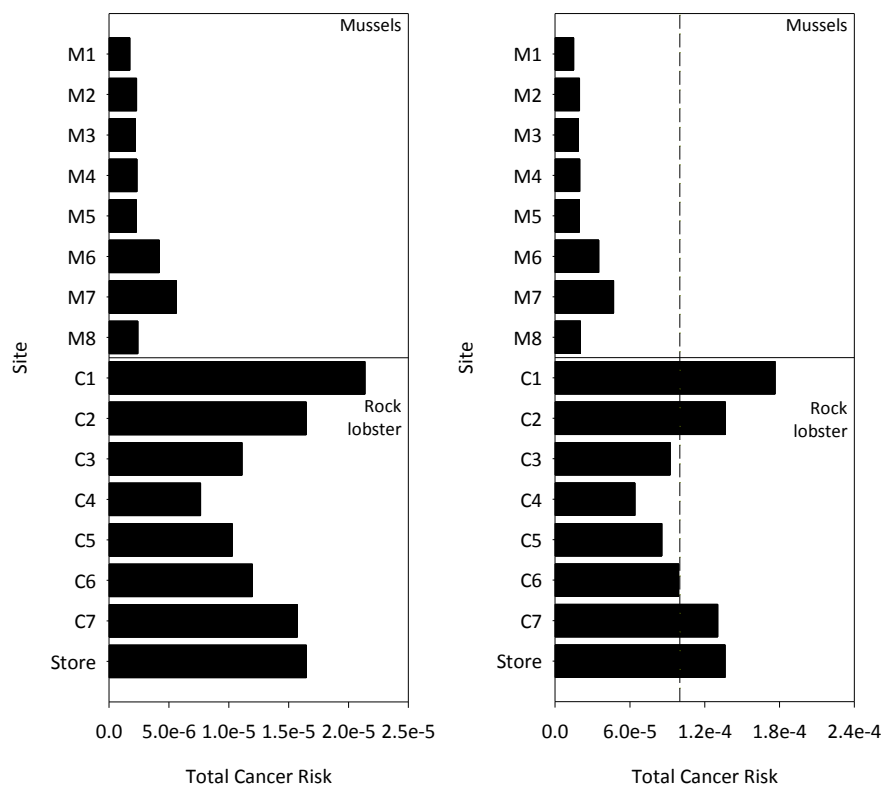


Figure 6.17. Total Cancer Risk for various chemicals in black mussels (*Mytilus galloprovincialis*) and west coast rock lobsters (*Jasus lalandii*) collected at sites along the Cape Peninsula area of Cape Town in 2016 as per the low-end (left) and high-end (right) consumer scenarios.

were considered in the risk assessment. Mussels and rock lobsters on the Cape Peninsula may or may not have other chemicals in their tissue at concentrations that pose a risk to the health of human consumers. If the former, then the risk may be higher than that calculated for this study.

Third, it was assumed consumers have historically and will in future consume mussels and rock lobsters collected from a particular site, and only that site at the stipulated consumption rate for the 30 year exposure period. This is unlikely as there are two 'no take' zones in the Table Mountain National Park Marine Protected Area (Karbonkelberg Sanctuary Zone and Cape of Good Hope Sanctuary Zone), and rock lobsters can only be collected recreationally for a short period of the year. For example, the rock lobster season for 2016/2017 totalled 21 days between 26 November 2016 - 17 April 2017. Since only four rock lobsters can be harvested per person per day a high-end consumer should theoretically not be able to harvest sufficient rock lobsters for their own consumption to exceed any appreciable health risk, even if they harvest at the maximum rate allowed.

Fourth, low- and high-end consumption rates may not apply to 'subsistence' consumers, that is, consumers that rely on fish and shellfish as a protein source more so than the 'average' person.

Last, and perhaps most importantly the ratio of inorganic to total arsenic in mussels and rock lobsters was assumed to be 2% due to the lack of empirical data on the ratio for these organisms. This ratio is higher than that reported for lobsters in other parts of the world. This creates uncertainty on the risk considering inorganic arsenic was the largest contributor to the estimated carcinogenic risk for rock lobsters. It is for this reason the City of Cape Town has initiated research to determine the ratio of inorganic to total arsenic in the tissue of mussels and rock lobsters collected along the Western Cape coastline.

It is important to note the calculated risks are estimates of current and future potential systemic and cancer risk. However, these do not predict health outcomes. Using approaches based on United States Environment Protection Agency guidance the potential risk is estimated in a conservative, public health protective manner that

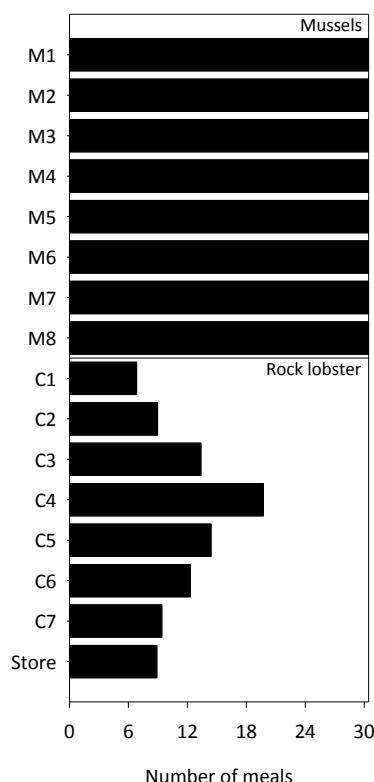


Figure 6.18. Number of meals of black mussels (*Mytilus galloprovincialis*) and west coast rock lobster (*Jasus lalandii*) collected at sites along the Cape Peninsula area of Cape Town in 2016 that can be consumed per month. The x-axis of the graphs is scaled to 30.44, that is, one meal per day.

usually tends to overestimate the risk, and actual health impacts are likely to be lower than the estimates.

Based on these uncertainties the findings of the risk assessment should not be construed as definitive but as an indicator of potential risk to the health of humans that consume mussels and rock lobsters collected on the Cape Peninsula.

6.4. Conclusions

Trends in the concentration of metals, polycyclic aromatic hydrocarbons, polychlorinated biphenyls and non-steroidal anti-inflammatory drugs in the tissue of black mussels and west coast rock lobsters collected at sites along the Atlantic seaboard of the Cape Peninsula in 2016 provided no evidence that

mussels or rock lobsters collected inshore of the outfalls had accumulated these chemicals to excessive concentrations in their tissue. This suggests effluent-derived constituents are not reaching nearshore and shoreline waters, or at least are not reaching these waters so often that mussels and rock lobsters are accumulating them excessively in their tissue. This conclusion is based primarily on the fact that non-steroidal anti-inflammatory drugs were not detected in the tissue of mussels and rock lobsters, as these are specific markers of effluent. Although mussels and/or rock lobsters had polycyclic aromatic hydrocarbons and/or polychlorinated in their tissue, these are often detected in mussels in other parts of the world since they are so widespread and their source could not be conclusively related to effluent discharge.

Assessment of potential systemic and cancer risks posed by metals, polycyclic aromatic hydrocarbons and polychlorinated biphenyls in the mussels and rock lobsters showed cadmium in mussels collected at one site poses a possible (but low) systemic risk to human health, while inorganic arsenic in rock lobsters collected at three sites and purchased in a store pose a possible (but low) cancer risk, but in both cases only for high-end consumers (*i.e.* those that eat a large amount of seafood on a daily basis). Eating a large amount of rock lobster over a sustained period may result in about two additional incidences of cancer per 10 000 members of the population. However, this estimate is based on the conservative assumption that the ratio of inorganic to total arsenic is 2%, which is higher than the ratio reported for lobsters in other parts of the world. If the ratio is assumed to be 0.2%, in line with the ratio reported for lobsters in other parts of the world, then inorganic arsenic in west coast rock lobsters does not pose a risk to human health. These findings will be reviewed once information on the ratio of inorganic to total arsenic has been established for black mussels and west coast rock lobster.